EXHIBIT 1

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Via Overnight Delivery
Division of Dockets Management
Food and Drug Administration
Department of Health and Human Services
5630 Fishers Lane
Room 1061 (HFA-305)
Rockville, Maryland 20852

CITIZEN PETITION

On behalf of Shire US, Inc. (Shire), the undersigned submits this petition under section 505 of the Federal Food, Drug, and Cosmetic Act (the FDC Act), 21 U.S.C. § 355, and 21 C.F.R. §§ 10.30 and 314.94, to request the Commissioner of Food and Drugs (hereinafter referred to as "FDA") to: (1) refrain from approving any abbreviated new drug application (ANDA) for Agrylin® (anagrelide hydrochloride) capsules that fails to include active metabolite monitoring in bioequivalency testing; and (2) require an ANDA applicant for anagrelide hydrochloride capsules to evaluate bioequivalence, monitoring the active metabolite under both fed and fasting conditions.

As explained below, Shire believes that FDA cannot approve an ANDA for Agrylin without the applicant demonstrating that its product results in comparable exposure to the reference listed drug's active metabolite, 3-hydroxy anagrelide, seen after Agrylin administration. Without such a demonstration, an ANDA applicant simply cannot unequivocally state that their product is bioequivalent to Agrylin.

A. Actions Requested

The undersigned requests that:

(1) FDA refrain from approving any ANDA for anagrelide hydrochloride capsules that fails to include active metabolite monitoring in bioequivalency testing. Specifically, FDA must require all ANDA applicants to monitor 3-hydroxy anagrelide in any bioequivalency study of anagrelide to ensure that a similar exposure to the active metabolite is achieved; and

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(2) FDA require any ANDA applicant for anagrelide hydrochloride capsules to monitor the active metabolite *under both fed and fasting conditions*, because it appears that food affects a patient's exposure to the parent drug (i.e., the active ingredient) and active metabolite in different ways.

FDA must reject an ANDA that fails to provide the aforementioned monitoring and related assurances of bioequivalence and to avoid potential safety and efficacy concerns, because the ANDA will fail to satisfy the statutory and regulatory requirements for approval. Without such assurances, the ANDA product cannot be considered to be the same as the innovator drug.

B. Statement of Grounds

1. Background

FDA approved a new drug application for Agrylin 0.5mg and 1mg capsules on March 14, 1997.[1] The active ingredient in Agrylin is anagrelide hydrochloride (6,7-Dichloro-1, 5-dihydroimidazo [2,1-b-]-quinazolin-2 (3H)-one hydrochloride monohydrate). Anagrelide hydrochloride is a highly potent and selective platelet lowering agent.

Agrylin is a prescription drug product indicated for the treatment of patients with thrombocythemia, a condition characterized by elevated blood platelets. Agrylin is used to reduce elevated platelet counts and the risk of thrombosis. Agrylin is also indicated for the amelioration of associated symptoms, including thrombo-hemorrhagic events.[2] FDA has designated Agrylin as the reference listed drug for anagrelide hydrochloride capsules.

Under the Orphan Drug Act, FDA granted Agrylin orphan drug exclusivity, which was set to expire on March 14, 2004. However, pursuant to section 505A of the FDC Act, 21 U.S.C. § 355a, and in accordance with FDA's Pediatric Written Request, Shire conducted a clinical trial that included a pediatric population, and obtained an additional six months of pediatric exclusivity for Agrylin. Therefore, Agrylin's exclusivity expires September 14, 2004.[3]

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[1] Roberts Pharmaceutical Corp. obtained the NDA approval and was subsequently acquired by Shire.

[2] A copy of the approved package insert is included as Attachment A.

[3] A copy of the relevant sections relating to Agrylin from FDA's Approved Drug Products with Therapeutic Equivalence Evaluations (commonly referred to as "The Orange Book") is Attachment B.

During Shire's recent clinical trial that included both adult and pediatric populations, the company established for the first time in the target patient population (i.e., essential thrombocythemia [ET] patients) the pharmacokinetics of the major metabolite of anagrelide, 3-hydroxy anagrelide. Earlier *in vitro* studies had demonstrated that the metabolite was equipotent with the parent molecule in its potential platelet lowering activity, and was forty times more potent as a phosphodiesterase III (PDE III) inhibitor and, thus, as a cardioactive agent. However, this patient study showed for the first time that plasma exposure to the active metabolite was more than twice that of the parent drug. The greater plasma exposure to the active metabolite indicates that the active metabolite is a major contributor to the efficacy and safety of Agrylin.

Shire also conducted a clinical pharmacokinetic study to investigate the effect of food on the disposition of the parent drug and active metabolite. The results of this study showed that food affected the pharmacokinetics of the active metabolite and the parent drug (anagrelide) differently.

On March 11, 2004, Shire submitted a supplemental NDA (sNDA) in response to the FDA's Pediatric Written Request. The sNDA included the aforementioned clinical study in pediatric and adult ET patients (and other myeloproliferative disorders (MPD)), and information on the 3-hydroxy anagrelide metabolite. Based on this submission, FDA's Division of Gastrointestinal and Coagulation Drug Products granted a 6-month extension of exclusivity for Agrylin. The sNDA's proposed pediatric labeling changes are still under review by the Division.

Shire submitted another sNDA on June 17, 2004, to incorporate additional changes to the labeling based on Phase 1 clinical interaction studies and information in Special Patient groups (i.e., renally and hepatically impaired subjects). The submitted information included data on the pharmacokinetics of 3-hydroxy anagrelide metabolite. The proposed labeling changes include amendments to the safety portions of the labeling (e.g., PRECAUTIONS, WARNINGS). The NDA review division has not yet completed its review of this sNDA.[4]

Shire has also notified FDA's Office of Generic Drugs (OGD) of the changes proposed to the labeling based on the metabolite, including those changes affecting the safety portions of the labeling. Additionally, Shire has provided OGD documentation justifying the monitoring of anagrelide's active metabolite in any bioequivalency study through controlled correspondence; the cover letter for this correspondence is included in Attachment C.

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[4] Shire considers the contents of the sNDAs, not yet approved by FDA, to be proprietary and confidential and not subject to public disclosure at this time. Therefore, we are not attaching them to this Citizen Petition.

2. Historical Context

The initial qualitative identification of 3-hydroxy anagrelide as a human metabolite of the drug occurred two years ago. However, difficulties experienced in the synthesis of the metabolite precluded the conduct, reporting, and submission of clinical pharmacokinetic data on the active metabolite to FDA until earlier this year.

Beginning in 2001, Shire conducted additional nonclinical and clinical studies on anagrelide to meet certain regulatory requirements of the European Medicines Agency (EMEA). It was during the course of these studies that Shire identified a new major metabolite of anagrelide. However, its subsequent chemical synthesis proved challenging. Initially, only small quantities of metabolite were available, enabling limited *in vitro* screening. In late 2002, the results of this screening showed the compound to be a highly potent PDEIII inhibitor and to possess comparable platelet-lowering potency to the parent drug.

However, the complexity of the chemical synthesis of the active metabolite delayed the availability of larger quantities of metabolite. Once larger quantities were available in early 2003, Shire initiated the conduct of *in vivo* whole animal studies to confirm, for example, the cardiovascular consequences of this PDEIII inhibition. In parallel, Shire established a clinical bioanalytical method for the metabolite, and clinical pharmacokinetic studies ensued.

As soon as practical, in third quarter 2003, Shire initiated analysis and reporting of the clinical pharmacokinetic study data. The company commenced the first patient study shortly thereafter. The above described data were necessary for Shire to fully determine the metabolite's contribution in man. Shire submitted results of the clinical study in adult and pediatric patients to FDA in the March 2004 sNDA.

In early 2004, Shire completed a definitive cardiovascular pharmacology study on anagrelide's active metabolite in the anesthetized dog model. The study demonstrated the qualitative comparability of the 3-hydroxy anagrelide to the reference positive inotrope milrinone, although 3-hydroxy anagrelide was 10-20 times more potent. Milrinone is a cardiostimulant used clinically in the treatment of congestive heart failure. However, such use requires careful monitoring in view of its profound effects on the cardiovascular system and consequential safety concerns. Similarly, monitoring of exposure to 3-hydroxy anagrelide from any new generic formulation of anagrelide should be undertaken to provide assurance that there is not an unexpected exposure to this cardiostimulant metabolite of anagrelide.

Shire has acted with due diligence. Despite the technical challenges relating to the chemical synthesis of the active metabolite, the company has provided FDA with the relevant clinical data on the metabolite as soon as practically possible.

3. Regulatory Overview

An ANDA must contain data demonstrating that the generic drug product is comparable or the "same as" the innovator drug product in dosage form, strength, route of administration, quality, performance characteristics and intended use. See 21 U.S.C. § 355(j)(2)(A); 21 C.F.R. § 314.94(a).[5] Therefore, an ANDA must contain the "same" active ingredient as the innovator drug and have essentially the same labeling. See 21 U.S.C. § 355(j)(2)(A). Further, a generic applicant must, with limited exception, scientifically show that its product is bioequivalent (i.e., performs in the same manner as the innovator drug). See 21 U.S.C. § 355(j)(2)(A)(iv). FDA will refuse to approve an ANDA if the information in the application "is insufficient to show that the drug is bioequivalent to the listed drug." 21 U.S.C. § 355(j)(4)(F).

According to the FDC Act, an ANDA drug product is bioequivalent to a listed drug [one listed in FDA's Orange Book] if:

> (i) the rate and extent of absorption of the drug do not show a significant difference from the rate and extent of absorption of the listed drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses; or
>
> (ii) the extent of absorption of the drug does not show a significant difference from the extent of absorption of the listed drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses and the difference from the listed drug in the rate of absorption of the drug is intentional, is reflected in its proposed labeling, is not essential to the attainment of effective body drug concentrations on chronic use, and is considered medically insignificant for the drug.

21 U.S.C. § 355(j)(8)(B); see also 21 C.F.R. § 320.1(e).[6]

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[5] All applicable statutory and regulatory citations are included in Attachment D.

[6] The statutory and regulatory conditions to demonstrate bioequivalence are not exclusive and do not preclude other means of establishing bioequivalence. When the above methods are not applicable, other *in vivo* or *in vitro* test methods to demonstrate bioequivalence may be appropriate. In some cases, FDA may waive the bioequivalence testing requirement. See 21 C.F.R. § 320.22.

FDA states that bioavailability may be measured or bioequivalence "may be demonstrated by several in vivo and in vitro methods." 21 C.F.R. § 320.24(a).[7] Applicants must conduct "bioavailability and bioequivalence testing using the most accurate, sensitive, and reproducible approach available" among those prescribed methods in the regulations. Id.

FDA lists, in relevant part, the following *in vivo* and *in vitro* approaches, in descending order of accuracy, sensitivity, and reproducibility, for determining the bioavailability or bioequivalence of a drug product:

> (1) (i) An in vivo test in humans in which the concentration of the active ingredient or active moiety, and, when appropriate, its active metabolite(s), in whole blood, plasma, serum, or other appropriate biological fluid is measured as a function of time. This approach is particularly applicable to dosage forms intended to deliver the active moiety to the bloodstream for systemic distribution within the body; or
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> (ii) An in vitro test that has been correlated with and is predictive of human in vivo bioavailability data; or
>
> (2) An in vivo test in humans in which the urinary excretion of the active moiety, and, when appropriate, its active metabolite(s), are measured as a function of time. The intervals at which measurements are taken should ordinarily be as short as possible so that the measure of the rate of elimination is as accurate as possible. Depending on the nature of the drug product, this approach may be applicable to the category of dosage forms described in paragraph (b)(1)(i) of this section. This method is not appropriate where urinary excretion is not a significant mechanism of elimination.
>
> (3) An in vivo test in humans in which an appropriate acute pharmacological effect of the active moiety, and, when appropriate, its active metabolite(s), are measured as a function of time if such effect can be measured with sufficient accuracy, sensitivity, and reproducibility. This approach is applicable to the category of dosage forms described in paragraph (b)(1)(i) of this section only when appropriate methods are not available for

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[7] The FDC Act defines "bioavailability" as "the rate and extent to which the active ingredient or therapeutic ingredient is absorbed from a drug and becomes available at the site of drug action." 21 U.S.C. § 355(j)(8)(A); see also 21 C.F.R. § 320.1(a).

> measurement of the concentration of the moiety, and, when appropriate, its active metabolite(s), in biological fluids or excretory products but a method is available for the measurement of an appropriate acute pharmacological effect. This approach may be particularly applicable to dosage forms that are not intended to deliver the active moiety to the bloodstream for systemic distribution.

21 C.F.R. § 320.24(b)(1)-(3).

FDA guidance states that applicants attempting to demonstrate bioequivalence to an active ingredient that is not highly soluble must do so using *in vivo* testing. See "Guidance for Industry: Waiver of In Vivo Bioavailability and Bioequivalence studies for Immediate-Release Solid Oral Dosage Forms Based on a Biopharmaceutics Classification System" (Aug. 2000). Attachment E. The standard *in vivo* bioequivalence study is conducted using a two-treatment crossover study design in a limited number of volunteers, typically 24-36 healthy adults. See The Orange Book, at ix. Attachment F. The study provides the rate of absorption, or bioavailability, of the generic drug, which the ANDA applicant can compare to the innovator drug.

Anagrelide is essentially unionized at physiological pH and is poorly water soluble (~1.5 ng/ml). Thus, according to FDA's guidance, anagrelide is not considered a "highly soluble" drug, because the maximum 1 mg dosage strength would not dissolve in the guideline-specified 250 ml of aqueous media. Therefore, a bioequivalence demonstration by a generic should be done using *in vivo* testing.

4. 3-Hydroxy Anagrelide is an Active Metabolite of Anagrelide Hydrochloride

Shire has demonstrated that 3-hydroxy anagrelide is an active metabolite of Agrylin capsules' active ingredient, anagrelide hydrochloride. Shire's recent studies show that 3-hydroxy anagrelide makes a meaningful contribution to the safety and efficacy of Agrylin. Furthermore, Shire's studies show that the active metabolite is generated during presystemic metabolism, which means that a patient's exposure to the active metabolite could be affected by changes in the drug formulation. The data supporting Shire's position in this Citizen Petition are summarized in Attachments G and H.

(a) Evidence for meaningful contribution of anagrelide's metabolite to the efficacy and safety of the drug

Anagrelide is extensively metabolized in man to two major metabolites; 3-hydroxy anagrelide and RL603. While RL603 has been shown to have no effect on megakaryocytopoeisis (and potentially, therefore, platelet lowering) (Erusalimsky, Hong,

and Franklin 2002), recent studies on 3-hydroxy anagrelide have revealed it to have significant and relevant pharmacology. Furthermore, results from a recent clinical pharmacokinetic study in ET and other MPD patients show plasma exposure to 3-hydroxy anagrelide exceeds plasma exposure to the parent drug (anagrelide) by 230%.

The effects of anagrelide and its metabolites on the differentiation of human $CD34^+$ stem cells into megakaryocytes, which ultimately give rise to blood platelets, was assessed using a well established model of megakaryocytopoeisis (Cohen-Solal 1997, Cramer 1997). All referenced articles are provided in Attachment I. RL603 was found to be inactive in this model (Erusalimsky, Hong and Franklin 2002). By contrast, Shire found that 3-hydroxy anagrelide has a comparable potency to the parent drug, as evidenced by a similar *in vitro* concentration resulting in 50% inhibition (IC_{50}) of megakaroycyte growth/differentiation. The most marked effect of the metabolite, like anagrelide, was on cell growth. Results from several Shire studies indicated mean IC_{50} values for effects on growth to be 27nM and compared to 48nM for effects on differentiation. (Attachment G, p. 7)

While anagrelide was already known to possess cardiovascular activity as the result of inhibition of PDEIII, the activity of its metabolites was only relatively recently determined. Shire found that anagrelide's active metabolite, 3-hydroxy anagrelide, was nearly forty times more potent as an inhibitor of PDEIII than anagrelide itself, having an *in vitro* concentration producing 50% inhibition (IC_{50}) of enzyme activity of 1.1nM. Shire confirmed these data in a second study that resulted in an average IC_{50} of 0.9 nM (Attachment G, p. 8). The second study also revealed that the other metabolite, RL603, was an extremely weak inhibitor of PDEIII, with an IC_{50} as high as 40,000nM.

Shire confirmed the expected *in vivo* cardiovascular activity of 3-hydroxy anagrelide in an anaesthetized dog model, where it was found to be 10-20 times more potent than the reference positive inotrope, milrinone, which it closely resembled with respect to its inotropic, chronotropic and vasodilatory activity. Thus, anagrelide's cardiostimulant activity, which manifests itself in side effects, such as tachycardia and palpitations, is likely due to the highly potent metabolite, 3-hydroxy anagrelide.

Therefore, based on the data summarized above and in Attachments G and H, Shire believes that the 3-hydroxy metabolite contributes significantly to Agrylin's pharmacological activity.

(b) Evidence for presystemic metabolism

A drug's formulation will only influence plasma exposure to an active metabolite when the metabolite is formed presystemically. As stated in recent FDA draft guidance, it is important to assess metabolite exposure in a bioequivalency study only when it is determined that the metabolite is formed presystemically. See "Draft Guidance for Industry: Bioavailability and Bioequivalence Studies for Orally Administered Drug Products – General Considerations" (July 2002); Attachment I.

Anagrelide has been shown to be quantitatively absorbed from the gastrointestinal (GI) tract, and is not chemically degraded within it. Therefore, anagrelide's oral bioavailability is determined only by presystemic metabolism. The absolute oral bioavailability of the drug will be a measure of the presystemic metabolism.

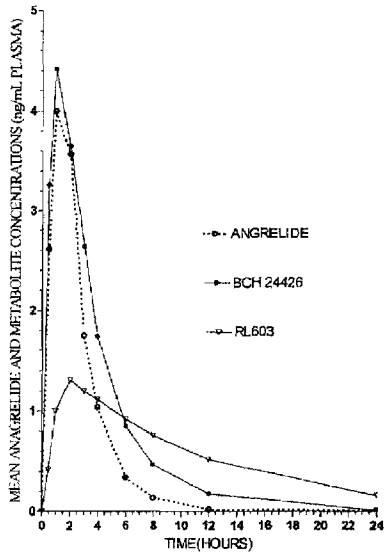
The unavailability of a clinical intravenous formulation of anagrelide has precluded the direct measurement of its absolute oral bioavailability. Nevertheless, other data provide valuable insight into the likelihood of presystemic metabolism of anagrelide.

(i) Data from healthy subjects (Attachment H, p. 5)

Shire conducted various clinical pharmacokinetic studies that have indirectly generated information on anagrelide's presystemic metabolism. Data from 38 healthy volunteers (age range 21-70, mean 52 years) who participated in three separate clinical pharmacokinetic studies demonstrated anagrelide to be rapidly absorbed (T_{max} of 1.3 hours).

Comparing the early concentration-time profiles of anagrelide and metabolite, the rate of formation of the 3-hydroxy anagrelide metabolite appeared to proceed in parallel with the absorption of the drug, suggesting that the metabolite's formation was effected by first-pass metabolism of anagrelide.

Figure 1: Plasma concentration time profile for anagrelide and its active metabolite, 3-hydroxy anagrelide (BCH24426), in man following a single 1 mg oral dose of the drug under fasting conditions



(ii) Studies in hepatically-impaired subjects (Attachment H, p. 5)

While gut wall or luminal metabolism can play a role, the most likely site of presystemic metabolism of a drug is the liver. An indication that the liver is involved in the biotransformation of anagrelide may be revealed by a comparison of anagrelide and metabolite exposure in normal and hepatically-impaired subjects.

Evidence that the liver plays a major role in the metabolism of anagrelide is supported by a pharmacokinetic study conducted in 10 subjects with moderate hepatic impairment. The study assessed the influence on the kinetics of both anagrelide and its metabolites. The study revealed an 8-fold increase in total exposure (AUC) to anagrelide. While part of this increase could be related to the 2.2-fold increase in half-life and a possible reduction in volume of distribution the increase also suggests that the liver plays a major role in the metabolism of anagrelide.

(iii) Oral clearance in the target patient population

Shire also recently investigated the pharmacokinetics of anagrelide and its metabolites in patients with ET and other MPDs. The study involved a comparison of younger patients (younger than 15 years of age) with a group of older patients.

Using the oral plasma clearance data, it is possible to estimate a drug's oral bioavailability. Furthermore, a drug's bioavailability is useful to measure a drug's presystemic metabolism, using the following equation:

$$F = 1/(1 + (CL_b/F)/Q_H)$$

Where F = bioavailability

CL_b/F = oral blood clearance

Q_H = liver blood flow (assume 1.35L/min)

(Rowland & Tozer 1995). The equation assumes complete oral absorption and clearance only by the liver (Attachment H, p.6-9).

Previous studies, specific to anagrelide, demonstrate that the above equation can be used to estimate anagrelide's oral bioavailability. Two human radiolabelled studies on orally-administered anagrelide have shown that between 75-80% of the administered radioactivity is recovered in the urine; implying near quantitative absorption (Gaver et al 1981 and Shire study). Furthermore, investigation of the possibility for presystemic gut floral or gut wall metabolism has been investigated and ruled out (Attachment H, p. 7).

A recent partitioning study has shown a plasma to blood ratio of 1.2:1. Applying this to the calculation of bioavailability in a group of 18 ET patients results in a mean +/- estimate of bioavailability of 52.6% +/- 12.5 (Attachment H, p.9). As a result, at least 48% of an orally administered dose is likely to be metabolized during the drug's initial passage through the liver (i.e., presystemically).

A more detailed account of the evidence supporting the belief that anagrelide's active metabolite is formed by presystemic hepatic metabolism is contained in Attachment H.

In summary, Shire's data supports the conclusion that the active metabolite of anagrelide, 3-hydroxy anagrelide, is formed by first-pass metabolism. As such, its formation could be influenced by changes in drug product formulation.

5. Food Intake Affects Exposure to Agrylin and its Active Metabolite in Different Ways

Shire proposes that bioequivalency studies on anagrelide be conducted under both fed and fasting conditions and, equally important, that such studies measure both the drug and its active metabolite.

A recent food interaction study conducted by Shire demonstrated that food affects exposure to anagrelide and the active metabolite in different ways. In the presence of food, a later T_{max} value for anagrelide was observed with a small reduction in C_{max} (~14%), but an overall increase in AUC of approximately 20%. On the other hand, the active metabolite, 3-hydroxy anagrelide, while again showing a later T_{max}, demonstrated a more pronounced reduction in C_{max} (~30%), but did not experience a change in the overall AUC. The observed changes with food indicate that the relationship between anagrelide and its active metabolite, with respect to their exposure-time profiles, is not straightforward and that bioequivalence studies on anagrelide should include monitoring of its active metabolite in both fed and fasting states.

6. FDA's Guidances to Industry and the Agency's Past Actions Require an ANDA Applicant to Provide Data Demonstrating Comparable Exposure to the Active Metabolite Between the Listed Drug and the Generic Version

Shire's requests in this Citizen Petition are consistent with FDA's guidances to industry and past agency determinations in similar situations where the parent drug's active metabolite could influence assessment of therapeutic equivalence of products. Specifically, an ANDA applicant must provide data demonstrating comparable exposure to the active metabolite between the listed drug and the generic version.

(a) General guidance

In the aforementioned Draft Guidance (July 2002), FDA stated, regarding metabolite monitoring in bioequivalency studies:

> For BE studies measurement of only parent drug released from the dosage form, rather than the metabolite, is generally recommended. The following are exceptions to this general approach:
>
> A metabolite may be formed as a result of gut wall or other presystemic metabolism. If the metabolite contributes meaningfully to safety and/or efficacy, the metabolite and the parent drug should be measured. When the relative activity of the metabolite is low and does not contribute meaningfully to safety and/or efficacy, it does not need to be measured. We recommend the parent drug measured in these BE studies be analyzed using the confidence interval approach. The metabolite data can be used to provide supportive evidence of

comparable therapeutic outcome.

Draft Guidance, at Section VI, paragraph B; Attachment J.

In this Citizen Petition, Shire has presented data that shows: (1) 3-hydroxy anagrelide is equipotent with its parent, anagrelide, as a platelet reducing agent, (2) 3-hydroxy anagrelide is up to forty times more potent than its parent as a cardiovascular agent, and (3) plasma exposure to the active metabolite exceeds the plasma exposure to anagrelide in the target patient population by a ratio of greater than two to one. Furthermore, evidence is provided for the metabolite's formation by first-pass metabolism. Thus, anagrelide's active metabolite would appear to meet FDA's own criteria for metabolite monitoring.

(b) Compound-specific guidance

For several compounds with active metabolites, FDA has issued specific guidance relating to the design of bioequivalence studies (e.g., tolmetin, guanabenz, selegiline, diltiazem and terfenadine), and FDA required their measurement. Guidance documents from Division of Bioequivalence, OGD, FDA; Attachment K. While no such guidance exists for anagrelide (information on the active metabolite has only just become available), the provision of such specific guidance for other compounds emphasizes the importance that FDA attaches to active metabolite monitoring.

7. Agrylin, Through Extensive Testing on Both Anagrelide and the Metabolite, is Safe and Effective, and Any ANDA Applicant for Anagrelide Must Conduct its own Testing to be Considered "The Same" as Agrylin Capsules

The FDC Act requires that a new drug be shown to be safe and effective. An ANDA product demonstrates that it is as safe and effective as the innovator drug by demonstrating that it is the "same as" the innovator, based on bioequivalence. Without data to demonstrate that the metabolite exposure of the ANDA applicant's product is comparable to the metabolite exposure from Agrylin, the ANDA applicant's product cannot be considered the "same as" the innovator's drug product.

Agrylin has undergone extensive testing to demonstrate that it is safe and effective. In the absence of the type of testing conducted by Shire and discussed in this Citizen Petition, an ANDA applicant will not be able to provide adequate assurances about any potential safety issues, such as the cardiovascular activity issue discussed above. Until such testing is performed, the ANDA applicant cannot demonstrate that its drug product is the "same as" Agrylin, such as the law requires. Although Shire will defer to FDA's

judgment on the type of testing that an ANDA applicant must perform, Shire believes that FDA must require ANDA applicants to perform such testing.

8. Conclusion

Under the FDC Act, FDA regulations, and relevant FDA guidance related to bioequivalence, a bioequivalency study of anagrelide hydrochloride must include specific data related to the active metabolite, 3-hydroxy anagrelide, and the effect of food on exposure of anagrelide and 3-hydroxy anagrelide. An ANDA for anagrelide hydrochloride that fails to include active metabolite monitoring *under fed and fasting testing conditions* cannot demonstrate meaningful bioequivalence and, thus, FDA must not approve any such ANDA.

For these reasons, FDA should require an ANDA applicant for anagrelide to monitor 3-hydroxy anagrelide in any bioequivalency study of anagrelide to ensure that a similar exposure to this active metabolite is achieved. In the absence of such assurances, an ANDA applicant cannot satisfy the statutory requirement of sameness to the innovator product.

C. Environmental Impact

As provided in 21 C.F.R. § 25.31, neither an environmental assessment nor an environmental impact statement is required.

D. Economic Impact

As provided in 21 C.F.R. § 10.30(b), economic impact information is to be submitted only when requested by the Commissioner following review of the petition.

E. Certification

The undersigned certifies, that to the best knowledge and belief of the undersigned, this petition includes all information and views on which the petition relies, and that it includes representative data and information known to the petitioner which are unfavorable to the petition.

Respectfully submitted,

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AGRYLIN®
(anagrelide hydrochloride)
Capsules
Rx only

DESCRIPTION

Name: AGRYLIN® (anagrelide hydrochloride)

Dosage Form: 0.5 mg and 1 mg capsules for oral administration

Active Ingredient: AGRYLIN® Capsules contain either 0.5 mg or 1 mg of anagrelide base (as anagrelide hydrochloride).

Inactive Ingredients: Anhydrous Lactose NF, Crospovidone NF, Lactose Monohydrate NF, Magnesium StearateNF, Microcrystalline Cellulose NF, Povidone USP.

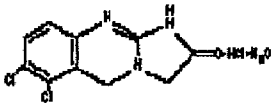
Pharmacological Classification: *Platelet-reducing agent.*

Chemical Name: 6,7-dichloro-1,5-dihydroimidazo[2,1-b]quinazolin-2(3H)-one monohydrochloride monohydrate.

Molecular formula: $C_{10}H_7Cl_2N_3O{\cdot}HCl{\cdot}H_2O$

Molecular weight: 310.55

Structural formula:



Appearance: Off-white powder.

Solubility: Water.....Very slightly soluble
Dimethyl Sulfoxide.....Sparingly soluble
Dimethylformamide.....*Sparingly soluble*

CLINICAL PHARMACOLOGY

The mechanism by which anagrelide reduces blood platelet count is still under investigation. Studies in patients support a hypothesis of dose-related reduction in platelet production resulting from a decrease in megakaryocyte hypermaturation. In blood withdrawn from normal volunteers treated with anagrelide, a disruption was found in the postmitotic phase of megakaryocyte development and a reduction in megakaryocyte size and ploidy. At therapeutic doses, anagrelide does not produce significant changes in white cell counts or coagulation parameters, and may have a small, but clinically insignificant effect on red cell parameters. Platelet aggregation is inhibited in people at doses higher than those required to reduce platelet count. Anagrelide inhibits cyclic AMP phosphodiesterase, as well as ADP- and collagen-induced platelet aggregation.

Following oral administration of ^{14}C-anagrelide in people, more than 70% of radioactivity was recovered in urine. Based on limited data, there appears to be a trend toward dose linearity between doses of 0.5 mg and 2.0 mg. At fasting and at a dose of 0.5 mg of anagrelide, the plasma half-life is 1.3 hours. The available plasma concentration time data at steady state in patients showed that anagrelide does not accumulate in plasma after repeated administration. The drug is extensively metabolized; less than 1% is recovered in the urine as anagrelide.

When a 0.5 mg dose of anagrelide was taken after food, its bioavailability (based on AUC values) was modestly reduced by an average of 13.8% and its plasma half-life slightly increased (to 1.8 hours), when compared with drug administered to the same subjects in the fasted state. The peak plasma level was lowered by an average of 45% and delayed by 2 hours.

CLINICAL STUDIES

A total of 942 patients with myeloproliferative disorders including 551 patients with Essential Thrombocythemia (ET), 117 patients with Polycythemia Vera (PV), 178 patients with Chronic Myelogenous Leukemia (CML), and 96 patients with other myeloproliferative disorders (OMPD), were treated with anagrelide in three clinical trials. Patients with OMPD included 87 patients who had Myeloid Metaplasia with Myelofibrosis (MMM), and 9 patients who had unknown myeloproliferative disorders.

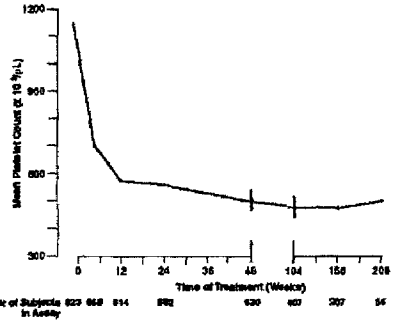
Clinical Studies

Patients with ET, PV, CML, or MMM were diagnosed based on the following criteria:

ET	PV*	MMM
• Platelet count ≥ 900,000/µL on two determinations • Profound megakaryocytic hyperplasia in bone marrow • Absence of Philadelphia chromosome • Normal red cell mass • Normal serum iron and ferritin and normal marrow iron stores CML • Persistent granulocyte count ≥ 50,000/µL without evidence of infection • Absolute basophil count ≥ 100/µL • Evidence for hyperplasia of the granulocytic line in the bone marrow • Philadelphia chromosome is present • Leukocyte alkaline phosphatase ≤ lower limit of the laboratory normal range	• A1 Increased red cell mass • A2 Normal arterial oxygen saturation • A3 Splenomegaly • B1 Platelet count ≥ 400,000/µL in absence of iron deficiency or bleeding • B2 Leukocytosis (≥ 12,000/µL, in the absence of infection) • B3 Elevated leukocyte alkaline phosphatase • B4 Elevated serum B_{12} † Diagnosis positive if A1, A2, and A3 present; or, if no splenomegaly, diagnosis is positive if A1 and A2 are present with any two of B1, B2, or B3.	• Myelofibrotic (hypocellular, fibrotic) bone marrow • Prominent megakaryocytic metaplasia in bone marrow • Splenomegaly • Moderate to severe normochromic normocytic anemia • White cell count may be variable; (80,000-100,000/µL) • Increased platelet count • Variable red cell mass; teardrop poikilocytes • Normal to high leukocyte alkaline phosphatase • Absence of Philadelphia chromosome

Patients were enrolled in clinical trials if their platelet count was ≥ 900,000/µL on two occasions or ≥ 650,000/µL on two occasions with documentation of symptoms associated with thrombocythemia. The mean duration of anagrelide therapy for ET, PV, CML, and OMPD patients was 65, 67, 40, and 44 weeks, respectively; 23% of patients received treatment for 2 years. Patients were treated with anagrelide starting at doses of 0.5-2.0 mg every 6 hours. The dose was increased if the platelet count was still high, but to no more than 12 mg each day. Efficacy was defined as reduction of platelet count to or near physiologic levels (150,000-400,000/µL). The criteria for defining subjects as "responders" were reduction in platelets for at least 4 weeks to ≤600,000/µL, or by at least 50% from baseline value. Subjects treated for less than 4 weeks were not considered evaluable. The results are depicted graphically below:

Patients with Thrombocytosis Secondary to Myeloproliferative Disorders: Mean Platelet Count During Anagrelide Therapy



		Time on Treatment						
		Weeks				Years		
	Baseline	4	12	24	48	2	3	4
Mean*	1131	683	575	526	484	460	437	457
N	923†	868	814	662	530	407	207	55

*x 10³/µL

† Nine hundred and forty-two subjects with myeloproliferative disorders were enrolled in three research studies. Of these, 923 had platelet counts over the duration of the studies.

AGRYLIN® was effective in phlebotomized patients as well as in patients treated with other concomitant therapies including hydroxyurea, aspirin, interferon, radioactive phosphorus, and alkylating agents.

INDICATIONS AND USAGE

AGRYLIN® Capsules are indicated for the treatment of patients with thrombocythemia, secondary to myeloproliferative disorders, to reduce the elevated platelet count and the risk of thrombosis and to ameliorate associated symptoms including thrombo-hemorrhagic events (see CLINICAL STUDIES, DOSAGE and ADMINISTRATION).

WARNINGS

Cardiovascular

Anagrelide should be used with caution in patients with known or suspected heart disease, and only if the potential benefits of therapy outweigh the potential risks. Because of the positive inotropic effects and side-effects of anagrelide, a pre-treatment cardiovascular examination is recommended along with careful monitoring during treatment. In humans, therapeutic doses of anagrelide may cause cardiovascular effects, including vasodilation, tachycardia, palpitations, and congestive heart failure.

Renal

It is recommended that patients with renal insufficiency (creatinine ≥ 2mg/dL) receive anagrelide when, in the physician's judgment, the potential benefits of therapy outweigh the potential risks. These patients should be monitored closely for signs of renal toxicity while receiving anagrelide (see ADVERSE REACTIONS, Urogenital System).

Hepatic

It is recommended that patients with evidence of hepatic dysfunction (bilirubin, SGOT, or measures of liver function >1.5 times the upper limit of normal) receive anagrelide when, in the physician's judgment, the potential benefits of therapy outweigh the potential risks. These patients should be monitored closely for signs of hepatic toxicity while receiving anagrelide (see *ADVERSE REACTIONS, Hepatic System*).

PRECAUTIONS

Laboratory Tests: Anagrelide therapy requires close clinical supervision of the patient. While the platelet count is being lowered (usually during the first two weeks of treatment), blood counts *(hemoglobin, white blood cells), liver function (SGOT, SGPT) and renal function (serum creatinine, BUN)* should be monitored.

In 9 subjects receiving a single 5 mg dose of anagrelide, standing blood pressure fell an average of 22/15 mm Hg, usually accompanied by dizziness. Only minimal changes in blood pressure were observed following a dose of 2 mg.

Cessation of AGRYLIN® Treatment: In general, interruption of anagrelide treatment is followed by an increase in platelet count. After sudden stoppage of anagrelide therapy, the increase in platelet count can be observed within four days.

Drug Interactions: Bioavailability studies evaluating possible interactions between anagrelide and other drugs have not been conducted. The most common medications used concomitantly with anagrelide have been aspirin, acetaminophen, furosemide, iron, ranitidine, hydroxyurea, and allopurinol. The most frequently used concomitant cardiac medication has been digoxin. Although drug-to-drug interaction studies have not been conducted, there is no clinical evidence to suggest that *anagrelide interacts with any of these compounds.*

There is a single case report which suggests that sucralfate may interfere with anagrelide absorption.

Food has no clinically significant effect on the bioavailability of anagrelide.

Carcinogenesis, Mutagenesis, Impairment of Fertility: *No long-term studies in animals have* been performed to evaluate carcinogenic potential of anagrelide hydrochloride. Anagrelide hydrochloride was not genotoxic in the Ames test, the mouse lymphoma cell (L5178Y, $TK^{+/-}$) forward mutation test, the human lymphocyte chromosome aberration test, or the mouse *micronucleus test.* Anagrelide hydrochloride at oral doses up to 240 mg/kg/day (3,440 mg/m²/day, 195 times the recommended maximum human dose based on body surface area) was found to have no effect on fertility and reproductive performance of male rats. However, in female rats, at oral doses of 80 mg/kg/day (360 mg/m²/day, 49 times the recommended maximum human dose based on body surface area) or higher, it disrupted implantation when administered in early pregnancy and retarded or blocked parturition when administered in late pregnancy.

Pregnancy: Pregnancy Category C.

(i) Teratogenic Effects

Teratology studies have been performed in pregnant rats at oral doses up to 900 mg/kg/day (5,400 mg/m²/day, 730 times the recommended maximum human dose based on body surface area) and in pregnant rabbits at oral doses up to 20 mg/kg/day

(240 mg/m²/day, 32 times the recommended maximum human dose based on body surface area) and have revealed no evidence of impaired fertility or harm to the fetus due to anagrelide hydrochloride.

(ii) Nongenotoxic Effects

A fertility and reproductive performance study performed in female rats revealed that anagrelide hydrochloride at oral doses of 60 mg/kg/day (360 mg/m²/day, 49 times the recommended maximum human dose based on body surface area) or higher disrupted implantation and exerted adverse effect on embryo/fetal survival.

A perinatal and postnatal study performed in female rats revealed that anagrelide hydrochloride at oral doses of 60 mg/kg/day (360 mg/m²/day, 49 times the recommended maximum human dose based on body surface area) or higher produced delay or blockage of parturition, deaths of nondelivering pregnant dams and their fully developed fetuses, and increased mortality in the pups born.

Five women became pregnant while on anagrelide treatment at doses of 1 to 4 mg/day. Treatment was stopped as soon as it was realized that they were pregnant. All delivered normal, healthy babies. There are no adequate and well-controlled studies in pregnant women. Anagrelide hydrochloride should be used during pregnancy only if the potential benefit justifies the potential risk to the fetus.

Anagrelide is not recommended in women who are or may become pregnant. If this drug is used during pregnancy, or if the patient becomes pregnant while taking this drug, the patient should be apprised of the potential harm to the fetus. Women of child-bearing potential should be instructed that they must not be pregnant and that they should use contraception while taking anagrelide. Anagrelide may cause fetal harm when administered to a pregnant woman.

Nursing Mothers: It is not known whether this drug is excreted in human milk. Because many drugs are excreted in human milk and because of the potential for serious adverse reaction in nursing infants from anagrelide hydrochloride, a decision should be made whether to discontinue nursing or to discontinue the drug, taking into account the importance of the drug to the mother.

Pediatric Use: The safety and efficacy of anagrelide in patients under the age of 18 years have not been established. Myeloproliferative disorders are uncommon in pediatric patients. Anagrelide has been used successfully in 12 pediatric patients (age range 6.8 to 17.4 years; 6 male and 6 female), including 8 patients with ET, 2 patients with CML, 1 patient with PV, and 1 patient with OMPD. Patients were started on therapy with 0.5 mg qid to a maximum daily dose of 10 mg. The median duration of treatment was 18.1 months with a range of 3.1 to 92 months. Three patients received treatment for greater than three years.

Geriatric Use: Of the total number of subjects in clinical studies of Agrylin, 42.1% were 65 years and over, while 14.9% were 75 years and over. No overall differences in safety or effectiveness were observed between these subjects and younger subjects, and other reported clinical experience has not identified differences in response between the elderly and younger patients, but greater sensitivity of some older individuals cannot be ruled out.

ADVERSE REACTIONS

Analysis of the adverse events in a population consisting of 942 patients diagnosed with myeloproliferative diseases of varying etiology (ET: 551; PV: 117; OMPD: 274) has shown that all disease groups have the same adverse event profile. While most reported adverse events during anagrelide therapy have been mild in intensity and have decreased in frequency with continued therapy, serious adverse events were reported in these patients. These include the following: congestive heart failure, myocardial infarction, cardiomyopathy, cardiomegaly, complete heart block, atrial fibrillation, cerebrovascular accident, pericarditis, pericardial effusion, pleural effusion, pulmonary infiltrates, pulmonary fibrosis, pulmonary hypertension, pancreatitis, gastric/duodenal ulceration, and seizure.

Of the 942 patients treated with anagrelide for a mean duration of approximately 65 weeks, 161 (17%) were discontinued from the study because of adverse events or abnormal laboratory test results. The most common adverse events for treatment discontinuation were headache, diarrhea, edema, palpitation, and abdominal pain. Overall, the occurrence rate of all adverse events was 17.9 per 1,000 treatment days. The occurrence rate of adverse events increased at higher dosages of anagrelide.

The most frequently reported adverse reactions to anagrelide (in 5% or greater of 942 patients with myeloproliferative disease) in clinical trials were:

Headache	43.5%
Palpitations	26.1%
Diarrhea	25.7%
Asthenia	23.1%
Edema, other	20.6%
Nausea	17.1%
Abdominal Pain	16.4%
Dizziness	15.4%
Pain, other	15.0%
Dyspnea	11.9%
Flatulence	10.2%
Vomiting	9.7%
Fever	8.9%
Peripheral Edema	8.5%
Rash, including urticaria	8.3%
Chest Pain	7.8%
Anorexia	7.7%
Tachycardia	7.5%
Pharyngitis	6.8%
Malaise	6.4%
Cough	6.3%
Paresthesia	5.9%
Back Pain	5.9%
Pruritus	5.5%
Dyspepsia	5.2%

Adverse events with an incidence of 1% to < 5% included:

Body as a Whole System: Flu symptoms, chills, photosensitivity.

Cardiovascular System: Arrhythmia, hemorrhage, hypertension, cardiovascular disease, angina pectoris, heart failure, postural hypotension, thrombosis, vasodilatation, migraine, syncope.

Digestive System: Constipation, GI distress, GI hemorrhage, gastritis, melena, aphthous stomatitis, eructation.

Hemic & Lymphatic System: Anemia, thrombocytopenia, ecchymosis, lymphadenopathy.

Platelet counts below 100,000/µL occurred in 84 patients (ET: 35; PV: 9; OMPD: 40), reduction below 50,000/µL occurred in 44 patients (ET: 7; PV: 6; OMPD: 31) while on anagrelide therapy. Thrombocytopenia promptly recovered upon discontinuation of anagrelide.

Hepatic System: Elevated liver enzymes were observed in 3 patients (ET: 2; OMPD: 1) during anagrelide therapy.

Musculoskeletal System: Arthralgia, myalgia, leg cramps.

Nervous System: Depression, somnolence, confusion, insomnia, nervousness, amnesia.

Nutritional Disorders: Dehydration.

Respiratory System: Rhinitis, epistaxis, respiratory disease, sinusitis, pneumonia, bronchitis, asthma.

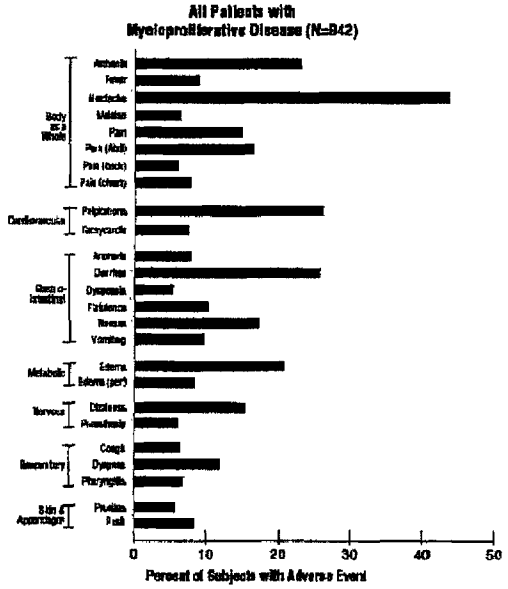
Skin and Appendages System: Skin disease, alopecia.

Special Senses: Amblyopia, abnormal vision, tinnitus, visual field abnormality, diplopia.

Urogenital System: Dysuria, hematuria.

Renal abnormalities occurred in 15 patients (ET: 10; PV: 4; OMPD: 1). Six ET, 4 PV and 1 with OMPD experienced renal failure (approximately 1%) while on anagrelide treatment; in 4 cases, the renal failure was considered to be possibly related to anagrelide treatment. The remaining 11 were found to have pre-existing renal impairment. Doses ranged from 1.5–6.0 mg/day, with exposure periods of 2 to 12 months. No dose adjustment was required because of renal insufficiency.

The adverse event profile for patients in clinical trials on anagrelide therapy (in 5% or greater of 942 patients with myeloproliferative diseases) is shown in the following bar graph:



OVERDOSAGE

Acute Toxicity and Symptoms

Single oral doses of anagrelide hydrochloride at 2,500, 1,500 and 200 mg/kg in mice, rats and monkeys, respectively, were not lethal. Symptoms of acute toxicity were: decreased motor activity in mice and rats and softened stools and decreased appetite in monkeys.

There are no reports of overdosage with anagrelide hydrochloride. Platelet reduction from anagrelide therapy is dose-related; therefore, thrombocytopenia, which can potentially cause bleeding, is expected from overdosage. Should overdosage occur, cardiac and central nervous system toxicity can also be expected.

Management and Treatment

In case of overdosage, close clinical supervision of the patient is required, this especially includes monitoring of the platelet count for thrombocytopenia. Dosage should be decreased or stopped, as appropriate, until the platelet count returns to within the normal range.

DOSAGE AND ADMINISTRATION

Treatment with **AGRYLIN®** Capsules should be initiated under close medical supervision. The recommended starting dosage of **AGRYLIN®** is 0.5 mg qid or 1 mg bid, which should be maintained for at least one week. Dosage should then be adjusted to the lowest effective dosage required to reduce and maintain platelet count below 600,000/µL, and ideally to the normal range. The dosage should be increased by not more than 0.5 mg/day in any one week. Dosage should not exceed 10 mg/day or 2.5 mg in a single dose (see PRECAUTIONS). The decision to treat asymptomatic young adults with essential thrombocythemia should be individualized. **There are no special requirements for dosing the geriatric population.**

To monitor the effect of anagrelide and prevent the occurrence of thrombocytopenia, platelet counts should be performed every two days during the first week of treatment and at least weekly thereafter until the maintenance dosage is reached.

Typically, platelet count begins to respond within 7 to 14 days at the proper dosage. The time to complete response, defined as platelet count ≤ 600,000/µL, ranged from 4 to 12 weeks. Most patients will experience an adequate response at a dose of 1.5 to 3.0 mg/day. Patients with known or suspected heart disease, renal insufficiency, or hepatic dysfunction should be monitored closely.

HOW SUPPLIED

AGRYLIN® is available as:

0.5 mg, opaque, white capsules (imprinted "Shire 063" in black ink.
NDC 54092-063-01 = bottle of 100

1 mg, opaque, gray capsules imprinted "Shire 064" in black ink
NDC 54092-064-01 = bottle of 100

Store at 25°C (77°F) excursions permitted to 15-30°C (59-86°F), in a light-resistant container. [See USP Controlled Room Temperature]

Manufactured for
Shire US Inc.
One Riverfront Place
Newport, KY 41071
By MALLINCKRODT INC.
Hobart, NY 13788

Rev. 4/03
Printed in USA

063 0117 012

B

Search results from the "OB_Rx" table for query on "020333."

Active Ingredient:	ANAGRELIDE HYDROCHLORIDE
Dosage Form;Route:	CAPSULE; ORAL
Proprietary Name:	AGRYLIN
Applicant:	SHIRE LABS
Strength:	EQ 0.5MG BASE
Application Number:	020333
Product Number:	001
Approval Date:	Mar 14, 1997
Reference Listed Drug	No
RX/OTC/DISCN:	RX
TE Code:	
Patent and Exclusivity Info for this product:	View

Active Ingredient:	ANAGRELIDE HYDROCHLORIDE
Dosage Form;Route:	CAPSULE; ORAL
Proprietary Name:	AGRYLIN
Applicant:	SHIRE LABS
Strength:	EQ 1MG BASE
Application Number:	020333
Product Number:	002
Approval Date:	Mar 14, 1997
Reference Listed Drug	Yes
RX/OTC/DISCN:	RX
TE Code:	
Patent and Exclusivity Info for this product:	View

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FDA/Center for Drug Evaluation and Research
Office of Generic Drugs
Division of Labeling and Program Support
Update Frequency:
Orange Book Data - **Monthly**
Orange Book Data Updated Through June, 2004
Orange Book Patent Data Only - **Daily**
Patent Data Last Updated: August 10, 2004

Patent and Exclusivity Search Results from query on Appl No 020333 Product 001 in the OB_Rx list.

Patent Data

There are no unexpired patents for this product in the Orange Book Database.

[Note: Title I of the 1984 Amendments does not apply to drug products submitted or approved under the former Section 507 of the Federal Food, Drug and Cosmetic Act (antibiotic products). Drug products of this category will not have patents listed.]

Exclusivity Data

Appl No	Prod No	Exclusivity Code	Exclusivity Expiration
020333	001	ODE	MAR 14,2004
020333	001	PED	SEP 14,2004

View a list of all patent use codes
View a list of all exclusivity codes

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Division of Labeling and Program Support
Update Frequency:
Orange Book Data - **Monthly**
Orange Book Data Updated Through June, 2004
Orange Book Patent Data Only - **Daily**
Patent Data Last Updated: August 10, 2004

C

Shire Pharmaceutical Development Inc
1801 Research Boulevard Suite 600 Rockville MD 20850 USA
Tel 240 453 6400 Fax 240 453 6404

30 June 2004

Shire

Gary Buehler
Division Director
Office of Generic Drugs (HFD-600)
Center for Drug Evaluation and Research
Food and Drug Administration
Metro Park North 2
7500 Standish Place
Rockville, MD 20855

Type of Submission:	**Controlled Correspondence / Request for Comment and Dialogue**
Re:	**NDA No. 20-333, Division of Gastroenterology and Coagulation Drug Products**
Product Name:	**AGRYLIN® (anagrelide hydrochloride)**

Dear Dr. Buehler:

With regard to the drug product Agrylin® (NDA 20-333), in accordance with the Office of Generic Drugs' (OGD) guidance on "controlled correspondence", Shire Pharmaceutical Development (Shire) is submitting a request for comment and dialogue with regard to monitoring of active metabolites in clinical bioequivalence studies. We are seeking OGD's guidance on the following question:

1. Does OGD agree that the active metabolite of anagrelide hydrochloride should be monitored in clinical bioequivalence studies?

Since the approval of Agrylin® in March 1997 by the FDA, Shire has progressed the development and approval of anagrelide in other international markets and this has resulted in further investigation and identification of the metabolites of anagrelide. The recent information on the role of the identified active metabolite, 3-hydroxy anagrelide, has been submitted to the FDA (Division of Gastroenterology and Coagulation Drug Products) as NDA supplements dated 12 March 2004 and 17 June 2004 to NDA 20-333 which included proposals to revise the product labeling.

For your information, a copy of the cover letter from the 12 March 2004 NDA supplement, and a full copy of the 17 June 2004 NDA supplement is included in this present "controlled correspondence" submission. The enclosed copy of the 17 June 2004 NDA supplement contains nonclinical and clinical pharmacokinetic data relevant to the active metabolite. It is Shire's opinion that the information presented and points raised in this letter and the supporting documents should be taken into consideration during the review of any pending anagrelide containing drug product ANDA submissions. We respectfully ask you to consider

a requirement for the measurement of the active metabolite of anagrelide in any clinical bioequivalence study.

As you are aware, the pediatric exclusivity period for Agrylin® expires on 11 September 2004, therefore, we look forward to the opportunity to discuss this issue with representatives of the OGD at your earliest convenience.

This "controlled correspondence" request is based on the following rationale:

- Recent nonclinical pharmacology and clinical pharmacokinetic studies have shown that the major metabolite of anagrelide, 3-hydroxy anagrelide, is considered to be responsible for most of the therapeutic platelet lowering activity and virtually all of the cardiovascular side effects associated with Agrylin®. Generation of this information was only possible following the successful chemical synthesis of 3-hydroxy anagrelide enabling the pharmacology and clinical pharmacokinetics of this metabolite to be examined.

- Clinical data (Shire study SPD422-202) indicate that the total plasma exposure (AUC, area under the plasma concentration-time curve) to this metabolite, 3-hydroxy anagrelide, in the target patient population exceeds that to anagrelide by 2.3:1. Note: the exposure to the metabolite is different in healthy volunteers.

- There is evidence that 3-hydroxy anagrelide is extensively (>50%) formed by first pass metabolism by CYP1A2 and that various extrinsic as well as intrinsic factors can significantly affect the amount of this active metabolite produced in vivo.

- A food interaction study (Shire study SPD422-109) suggests that the parent drug, anagrelide, may not necessarily act as a surrogate for the pharmacokinetic behaviour of the active metabolite 3-hydroxy anagrelide. While food significantly increased the AUC for anagrelide, it had no effect on the extent of exposure to the active metabolite. Furthermore food depressed the C_{max} for the anagrelide twice as much as that of the active metabolite.

It is the opinion of Shire that such a request for the monitoring of the active metabolite is consistent with the current FDA guidance on the conduct of bioequivalence studies.

A "controlled correspondence" supporting document is enclosed which provides a justification that monitoring of anagrelide's active metabolite, 3-hydroxy anagrelide, in clinical bioequivalence studies should be considered by the OGD. The document consists of an overview of the contribution of the active metabolite, 3-hydroxy anagrelide to the safety and efficacy of the drug product. The overview is supported by two appendices that provide more detail on the pharmacology, pharmacokinetics and evidence of pre-systemic metabolism of anagrelide.

You will have received a copy of a letter (dated 17 June 2004) sent to Dr. Robert Justice at the Division of Gastroenterology and Coagulation Drug Products that accompanied the recent labelling supplement to the Agrylin NDA. As mentioned earlier, a copy of the NDA supplement is included in this request for your information (pertinent clinical information is provided by the following studies: SPD422-103 renal impairment, SPD422-104 hepatic impairment, SPD422-107 aspirin interaction, SPD422-109 food effect, SPD422-202 clinical paharmacokinetic study in patients).

I look forward to hearing from you in the near future regarding the active metabolite question posed above. If deemed appropriate, Shire would welcome the opportunity to meet with representatives of OGD to further discuss the issues.

Should you have any questions, please do not hesitate to contact me at 240-453-6442 (phone) or 240-453-6456 (fax) or via email at csymington@us.shire.com.

Kind regards,

Catherine N. Symington
Senior Manager, Regulatory Affairs

Enclosures:

1. Supporting Documentation to justify metabolite monitoring
2. 12 March 2004 NDA supplement addressed to Division of Gastroenterology and Coagulation Drug Products (copy of cover letter)
3. 17 June 2004 NDA supplement addressed to Division of Gastroenterology and Coagulation Drug Products (full copy)

D

TITLE 21--FOOD AND DRUGS

CHAPTER 9--FEDERAL FOOD, DRUG, AND COSMETIC ACT

SUBCHAPTER V--DRUGS AND DEVICES

Part A--Drugs and Devices

Sec. 355. New drugs

(a) Necessity of effective approval of application

No person shall introduce or deliver for introduction into interstate commerce any new drug, unless an approval of an application filed pursuant to subsection (b) or (j) of this section is effective with respect to such drug.

(b) Filing application; contents

(1) Any person may file with the Secretary an application with respect to any drug subject to the provisions of subsection (a) of this section. Such person shall submit to the Secretary as a part of the application (A) full reports of investigations which have been made to show whether or not such drug is safe for use and whether such drug is effective in use; (B) a full list of the articles used as components of such drug; (C) a full statement of the composition of such drug; (D) a full description of the methods used in, and the facilities and controls used for, the manufacture, processing, and packing of such drug; (E) such samples of such drug and of the articles used as components thereof as the Secretary may require; and (F) specimens of the labeling proposed to be used for such drug. The applicant shall file with the application the patent number and the expiration date of any patent which claims the drug for which the applicant submitted the application or which claims a method of using such drug and with respect to which a claim of patent infringement could reasonably be asserted if a person not licensed by the owner engaged in the manufacture, use, or sale of the drug. If an application is filed under this subsection for a drug and a patent which claims such drug or a method of using such drug is issued after the filing date but before approval of the application, the applicant shall amend the application to include the information required by the preceding sentence. Upon approval of the application, the Secretary shall publish information submitted under the two preceding sentences. The Secretary shall, in consultation with the Director of the National Institutes of Health and with representatives of the drug manufacturing industry, review and develop guidance, as appropriate, on the inclusion of women and minorities in clinical trials required by clause (A).

(2) An application submitted under paragraph (1) for a drug for which the investigations described in clause (A) of such paragraph and relied upon by the applicant for approval of the application were not conducted by or for the applicant and for which the applicant has not obtained a right of reference or use from the person by or for whom the

investigations were conducted shall also include--

(A) a certification, in the opinion of the applicant and to the best of his knowledge, with respect to each patent which claims the drug for which such investigations were conducted or which claims a use for such drug for which the applicant is seeking approval under this subsection and for which information is required to be filed under paragraph (1) or subsection (c) of this section--

(i) that such patent information has not been filed,

(ii) that such patent has expired,

(iii) of the date on which such patent will expire, or

(iv) that such patent is invalid or will not be infringed by the manufacture, use, or sale of the new drug for which the application is submitted; and

(B) if with respect to the drug for which investigations described in paragraph (1)(A) were conducted information was filed under paragraph (1) or subsection (c) of this section for a method of use patent which does not claim a use for which the applicant is seeking approval under this subsection, a statement that the method of use patent does not claim such a use.

(3)(A) An applicant who makes a certification described in paragraph (2)(A)(iv) shall include in the application a statement that the applicant will give the notice required by subparagraph (B) to--

(i) each owner of the patent which is the subject of the certification or the representative of such owner designated to receive such notice, and

(ii) the holder of the approved application under subsection (b) of this section for the drug which is claimed by the patent or a use of which is claimed by the patent or the representative of such holder designated to receive such notice.

(B) The notice referred to in subparagraph (A) shall state that an application has been submitted under this subsection for the drug with respect to which the certification is made to obtain approval to engage in the commercial manufacture, use, or sale of the drug before the expiration of the patent referred to in the certification. Such notice shall include a detailed statement of the factual and legal basis of the applicant's opinion that the patent is not valid or will not be infringed.

(C) If an application is amended to include a certification described in paragraph (2)(A)(iv), the notice required by subparagraph (B) shall be given when the amended application is submitted.

(4)(A) The Secretary shall issue guidance for the individuals who review applications submitted under paragraph (1) or under section 262 of title 42, which shall relate to promptness in conducting the review, technical excellence, lack of bias and conflict of interest, and knowledge of regulatory and scientific standards, and which shall apply equally to all individuals who review such applications.

(B) The Secretary shall meet with a sponsor of an investigation or an applicant for approval for a drug under this subsection or section 262 of title 42 if the sponsor or applicant makes a reasonable written request for a meeting for the purpose of reaching agreement on the design and size of clinical trials intended to form the primary basis of an effectiveness claim. The sponsor or applicant shall provide information necessary for discussion and agreement on the design and size of the clinical trials. Minutes of any such meeting shall be prepared by the Secretary and made available to the sponsor or applicant upon request.

(C) Any agreement regarding the parameters of the design and size of

clinical trials of a new drug under this paragraph that is reached between the Secretary and a sponsor or applicant shall be reduced to writing and made part of the administrative record by the Secretary. Such agreement shall not be changed after the testing begins, except--

(i) with the written agreement of the sponsor or applicant; or

(ii) pursuant to a decision, made in accordance with subparagraph (D) by the director of the reviewing division, that a substantial scientific issue essential to determining the safety or effectiveness of the drug has been identified after the testing has begun.

(D) A decision under subparagraph (C)(ii) by the director shall be in writing and the Secretary shall provide to the sponsor or applicant an opportunity for a meeting at which the director and the sponsor or applicant will be present and at which the director will document the scientific issue involved.

(E) The written decisions of the reviewing division shall be binding upon, and may not directly or indirectly be changed by, the field or compliance division personnel unless such field or compliance division personnel demonstrate to the reviewing division why such decision should be modified.

(F) No action by the reviewing division may be delayed because of the unavailability of information from or action by field personnel unless the reviewing division determines that a delay is necessary to assure the marketing of a safe and effective drug.

(G) For purposes of this paragraph, the reviewing division is the division responsible for the review of an application for approval of a drug under this subsection or section 262 of title 42 (including all scientific and medical matters, chemistry, manufacturing, and controls).

(c) Period for approval of application; period for, notice, and expedition of hearing; period for issuance of order

(1) Within one hundred and eighty days after the filing of an application under subsection (b) of this section, or such additional period as may be agreed upon by the Secretary and the applicant, the Secretary shall either--

(A) approve the application if he then finds that none of the grounds for denying approval specified in subsection (d) of this section applies, or

(B) give the applicant notice of an opportunity for a hearing before the Secretary under subsection (d) of this section on the question whether such application is approvable. If the applicant elects to accept the opportunity for hearing by written request within thirty days after such notice, such hearing shall commence not more than ninety days after the expiration of such thirty days unless the Secretary and the applicant otherwise agree. Any such hearing shall thereafter be conducted on an expedited basis and the Secretary's order thereon shall be issued within ninety days after the date fixed by the Secretary for filing final briefs.

(2) If the patent information described in subsection (b) of this section could not be filed with the submission of an application under subsection (b) of this section because the application was filed before the patent information was required under subsection (b) of this section or a patent was issued after the application was approved under such subsection, the holder of an approved application shall file with the Secretary the patent number and the expiration date of any patent which claims the drug for which the application was submitted or which claims a method of using such drug and with respect to which a claim of patent

infringement could reasonably be asserted if a person not licensed by the owner engaged in the manufacture, use, or sale of the drug. If the holder of an approved application could not file patent information under subsection (b) of this section because it was not required at the time the application was approved, the holder shall file such information under this subsection not later than thirty days after September 24, 1984, and if the holder of an approved application could not file patent information under subsection (b) of this section because no patent had been issued when an application was filed or approved, the holder shall file such information under this subsection not later than thirty days after the date the patent involved is issued. Upon the submission of patent information under this subsection, the Secretary shall publish it.

(3) The approval of an application filed under subsection (b) of this section which contains a certification required by paragraph (2) of such subsection shall be made effective on the last applicable date determined under the following:

(A) If the applicant only made a certification described in clause (i) or (ii) of subsection (b)(2)(A) of this section or in both such clauses, the approval may be made effective immediately.

(B) If the applicant made a certification described in clause (iii) of subsection (b)(2)(A) of this section, the approval may be made effective on the date certified under clause (iii).

(C) If the applicant made a certification described in clause (iv) of subsection (b)(2)(A) of this section, the approval shall be made effective immediately unless an action is brought for infringement of a patent which is the subject of the certification before the expiration of forty-five days from the date the notice provided under paragraph (3)(B) is received. If such an action is brought before the expiration of such days, the approval may be made effective upon the expiration of the thirty-month period beginning on the date of the receipt of the notice provided under paragraph (3)(B) or such shorter or longer period as the court may order because either party to the action failed to reasonably cooperate in expediting the action, except that--

(i) if before the expiration of such period the court decides that such patent is invalid or not infringed, the approval may be made effective on the date of the court decision,

(ii) if before the expiration of such period the court decides that such patent has been infringed, the approval may be made effective on such date as the court orders under section 271(e)(4)(A) of title 35, or

(iii) if before the expiration of such period the court grants a preliminary injunction prohibiting the applicant from engaging in the commercial manufacture or sale of the drug until the court decides the issues of patent validity and infringement and if the court decides that such patent is invalid or not infringed, the approval shall be made effective on the date of such court decision.

In such an action, each of the parties shall reasonably cooperate in expediting the action. Until the expiration of forty-five days from the date the notice made under paragraph (3)(B) is received, no action may be brought under section 2201 of title 28 for a declaratory judgment with respect to the patent. Any action brought under such section 2201 shall be brought in the judicial district where the defendant has its principal place of business or a regular and established place of business.

(D)(i) If an application (other than an abbreviated new drug

application) submitted under subsection (b) of this section for a drug, no active ingredient (including any ester or salt of the active ingredient) of which has been approved in any other application under subsection (b) of this section, was approved during the period beginning January 1, 1982, and ending on September 24, 1984, the Secretary may not make the approval of another application for a drug for which the investigations described in clause (A) of subsection (b)(1) of this section and relied upon by the applicant for approval of the application were not conducted by or for the applicant and for which the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted effective before the expiration of ten years from the date of the approval of the application previously approved under subsection (b) of this section.

(ii) If an application submitted under subsection (b) of this section for a drug, no active ingredient (including any ester or salt of the active ingredient) of which has been approved in any other application under subsection (b) of this section, is approved after September 24, 1984, no application which refers to the drug for which the subsection (b) application was submitted and for which the investigations described in clause (A) of subsection (b)(1) of this section and relied upon by the applicant for approval of the application were not conducted by or for the applicant and for which the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted may be submitted under subsection (b) of this section before the expiration of five years from the date of the approval of the application under subsection (b) of this section, except that such an application may be submitted under subsection (b) of this section after the expiration of four years from the date of the approval of the subsection (b) application if it contains a certification of patent invalidity or noninfringement described in clause (iv) of subsection (b)(2)(A) of this section. The approval of such an application shall be made effective in accordance with this paragraph except that, if an action for patent infringement is commenced during the one-year period beginning forty-eight months after the date of the approval of the subsection (b) application, the thirty-month period referred to in subparagraph (C) shall be extended by such amount of time (if any) which is required for seven and one-half years to have elapsed from the date of approval of the subsection (b) application.

(iii) If an application submitted under subsection (b) of this section for a drug, which includes an active ingredient (including any ester or salt of the active ingredient) that has been approved in another application approved under subsection (b) of this section, is approved after September 24, 1984, and if such application contains reports of new clinical investigations (other than bioavailability studies) essential to the approval of the application and conducted or sponsored by the applicant, the Secretary may not make the approval of an application submitted under subsection (b) of this section for the conditions of approval of such drug in the approved subsection (b) application effective before the expiration of three years from the date of the approval of the application under subsection (b) of this section if the investigations described in clause (A) of subsection (b)(1) of this section and relied upon by the applicant for approval of the application were not conducted by or for the applicant and if the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted.

(iv) If a supplement to an application approved under subsection (b) of this section is approved after September 24, 1984, and the

supplement contains reports of new clinical investigations (other than bioavailabilty \1\ studies) essential to the approval of the supplement and conducted or sponsored by the person submitting the supplement, the Secretary may not make the approval of an application submitted under subsection (b) of this section for a change approved in the supplement effective before the expiration of three years from the date of the approval of the supplement under subsection (b) of this section if the investigations described in clause (A) of subsection (b)(1) of this section and relied upon by the applicant for approval of the application were not conducted by or for the applicant and if the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted.

\1\ So in original. Probably should be ``bioavailability''.

(v) If an application (or supplement to an application) submitted under subsection (b) of this section for a drug, which includes an active ingredient (including any ester or salt of the active ingredient) that has been approved in another application under subsection (b) of this section, was approved during the period beginning January 1, 1982, and ending on September 24, 1984, the Secretary may not make the approval of an application submitted under this subsection and for which the investigations described in clause (A) of subsection (b)(1) of this section and relied upon by the applicant for approval of the application were not conducted by or for the applicant and for which the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted and which refers to the drug for which the subsection (b) application was submitted effective before the expiration of two years from September 24, 1984.

(4) A drug manufactured in a pilot or other small facility may be used to demonstrate the safety and effectiveness of the drug and to obtain approval for the drug prior to manufacture of the drug in a larger facility, unless the Secretary makes a determination that a full scale production facility is necessary to ensure the safety or effectiveness of the drug.

(d) Grounds for refusing application; approval of application; ``substantial evidence'' defined

If the Secretary finds, after due notice to the applicant in accordance with subsection (c) of this section and giving him an opportunity for a hearing, in accordance with said subsection, that (1) the investigations, reports of which are required to be submitted to the Secretary pursuant to subsection (b) of this section, do not include adequate tests by all methods reasonably applicable to show whether or not such drug is safe for use under the conditions prescribed, recommended, or suggested in the proposed labeling thereof; (2) the results of such tests show that such drug is unsafe for use under such conditions or do not show that such drug is safe for use under such conditions; (3) the methods used in, and the facilities and controls used for, the manufacture, processing, and packing of such drug are inadequate to preserve its identity, strength, quality, and purity; (4) upon the basis of the information submitted to him as part of the application, or upon the basis of any other information before him with respect to such drug, he has insufficient information to determine whether such drug is safe for use under such conditions; or (5) evaluated on the basis of the information submitted to him as part of

the application and any other information before him with respect to such drug, there is a lack of substantial evidence that the drug will have the effect it purports or is represented to have under the conditions of use prescribed, recommended, or suggested in the proposed labeling thereof; or (6) the application failed to contain the patent information prescribed by subsection (b) of this section; or (7) based on a fair evaluation of all material facts, such labeling is false or misleading in any particular; he shall issue an order refusing to approve the application. If, after such notice and opportunity for hearing, the Secretary finds that clauses (1) through (6) do not apply, he shall issue an order approving the application. As used in this subsection and subsection (e) of this section, the term ``substantial evidence'' means evidence consisting of adequate and well-controlled investigations, including clinical investigations, by experts qualified by scientific training and experience to evaluate the effectiveness of the drug involved, on the basis of which it could fairly and responsibly be concluded by such experts that the drug will have the effect it purports or is represented to have under the conditions of use prescribed, recommended, or suggested in the labeling or proposed labeling thereof. If the Secretary determines, based on relevant science, that data from one adequate and well-controlled clinical investigation and confirmatory evidence (obtained prior to or after such investigation) are sufficient to establish effectiveness, the Secretary may consider such data and evidence to constitute substantial evidence for purposes of the preceding sentence.

(e) Withdrawal of approval; grounds; immediate suspension upon finding imminent hazard to public health

The Secretary shall, after due notice and opportunity for hearing to the applicant, withdraw approval of an application with respect to any drug under this section if the Secretary finds (1) that clinical or other experience, tests, or other scientific data show that such drug is unsafe for use under the conditions of use upon the basis of which the application was approved; (2) that new evidence of clinical experience, not contained in such application or not available to the Secretary until after such application was approved, or tests by new methods, or tests by methods not deemed reasonably applicable when such application was approved, evaluated together with the evidence available to the Secretary when the application was approved, shows that such drug is not shown to be safe for use under the conditions of use upon the basis of which the application was approved; or (3) on the basis of new information before him with respect to such drug, evaluated together with the evidence available to him when the application was approved, that there is a lack of substantial evidence that the drug will have the effect it purports or is represented to have under the conditions of use prescribed, recommended, or suggested in the labeling thereof; or (4) the patent information prescribed by subsection (c) of this section was not filed within thirty days after the receipt of written notice from the Secretary specifying the failure to file such information; or (5) that the application contains any untrue statement of a material fact: Provided, That if the Secretary (or in his absence the officer acting as Secretary) finds that there is an imminent hazard to the public health, he may suspend the approval of such application immediately, and give the applicant prompt notice of his action and afford the applicant the opportunity for an expedited hearing under this subsection; but the authority conferred by this proviso to suspend the approval of an application shall not be delegated. The Secretary may also, after due notice and opportunity for hearing to the applicant, withdraw the approval of an application submitted under subsection (b) or (j) of this

section with respect to any drug under this section if the Secretary finds (1) that the applicant has failed to establish a system for maintaining required records, or has repeatedly or deliberately failed to maintain such records or to make required reports, in accordance with a regulation or order under subsection (k) of this section or to comply with the notice requirements of section 360(k)(2) of this title, or the applicant has refused to permit access to, or copying or verification of, such records as required by paragraph (2) of such subsection; or (2) that on the basis of new information before him, evaluated together with the evidence before him when the application was approved, the methods used in, or the facilities and controls used for, the manufacture, processing, and packing of such drug are inadequate to assure and preserve its identity, strength, quality, and purity and were not made adequate within a reasonable time after receipt of written notice from the Secretary specifying the matter complained of; or (3) that on the basis of new information before him, evaluated together with the evidence before him when the application was approved, the labeling of such drug, based on a fair evaluation of all material facts, is false or misleading in any particular and was not corrected within a reasonable time after receipt of written notice from the Secretary specifying the matter complained of. Any order under this subsection shall state the findings upon which it is based.

(f) Revocation of order refusing, withdrawing or suspending approval of application

Whenever the Secretary finds that the facts so require, he shall revoke any previous order under subsection (d) or (e) of this section refusing, withdrawing, or suspending approval of an application and shall approve such application or reinstate such approval, as may be appropriate.

(g) Service of orders

Orders of the Secretary issued under this section shall be served (1) in person by any officer or employee of the department designated by the Secretary or (2) by mailing the order by registered mail or by certified mail addressed to the applicant or respondent at his last-known address in the records of the Secretary.

(h) Appeal from order

An appeal may be taken by the applicant from an order of the Secretary refusing or withdrawing approval of an application under this section. Such appeal shall be taken by filing in the United States court of appeals for the circuit wherein such applicant resides or has his principal place of business, or in the United States Court of Appeals for the District of Columbia Circuit, within sixty days after the entry of such order, a written petition praying that the order of the Secretary be set aside. A copy of such petition shall be forthwith transmitted by the clerk of the court to the Secretary, or any officer designated by him for that purpose, and thereupon the Secretary shall certify and file in the court the record upon which the order complained of was entered, as provided in section 2112 of title 28. Upon the filing of such petition such court shall have exclusive jurisdiction to affirm or set aside such order, except that until the filing of the record the Secretary may modify or set aside his order. No objection to the order of the Secretary shall be considered by the court unless such objection shall have been urged before the Secretary or unless there were reasonable grounds for failure so to do. The finding of the Secretary as

to the facts, if supported by substantial evidence, shall be conclusive. If any person shall apply to the court for leave to adduce additional evidence, and shall show to the satisfaction of the court that such additional evidence is material and that there were reasonable grounds for failure to adduce such evidence in the proceeding before the Secretary, the court may order such additional evidence to be taken before the Secretary and to be adduced upon the hearing in such manner and upon such terms and conditions as to the court may seem proper. The Secretary may modify his findings as to the facts by reason of the additional evidence so taken, and he shall file with the court such modified findings which, if supported by substantial evidence, shall be conclusive, and his recommendation, if any, for the setting aside of the original order. The judgment of the court affirming or setting aside any such order of the Secretary shall be final, subject to review by the Supreme Court of the United States upon certiorari or certification as provided in section 1254 of title 28. The commencement of proceedings under this subsection shall not, unless specifically ordered by the court to the contrary, operate as a stay of the Secretary's order.

(i) Exemptions of drugs for research; discretionary and mandatory conditions; direct reports to Secretary

(1) The Secretary shall promulgate regulations for exempting from the operation of the foregoing subsections of this section drugs intended solely for investigational use by experts qualified by scientific training and experience to investigate the safety and effectiveness of drugs. Such regulations may, within the discretion of the Secretary, among other conditions relating to the protection of the public health, provide for conditioning such exemption upon--

(A) the submission to the Secretary, before any clinical testing of a new drug is undertaken, of reports, by the manufacturer or the sponsor of the investigation of such drug, of preclinical tests (including tests on animals) of such drug adequate to justify the proposed clinical testing;

(B) the manufacturer or the sponsor of the investigation of a new drug proposed to be distributed to investigators for clinical testing obtaining a signed agreement from each of such investigators that patients to whom the drug is administered will be under his personal supervision, or under the supervision of investigators responsible to him, and that he will not supply such drug to any other investigator, or to clinics, for administration to human beings;

(C) the establishment and maintenance of such records, and the making of such reports to the Secretary, by the manufacturer or the sponsor of the investigation of such drug, of data (including but not limited to analytical reports by investigators) obtained as the result of such investigational use of such drug, as the Secretary finds will enable him to evaluate the safety and effectiveness of such drug in the event of the filing of an application pursuant to subsection (b) of this section; and

(D) the submission to the Secretary by the manufacturer or the sponsor of the investigation of a new drug of a statement of intent regarding whether the manufacturer or sponsor has plans for assessing pediatric safety and efficacy.

(2) Subject to paragraph (3), a clinical investigation of a new drug may begin 30 days after the Secretary has received from the manufacturer or sponsor of the investigation a submission containing such information about the drug and the clinical investigation, including--

(A) information on design of the investigation and adequate

reports of basic information, certified by the applicant to be accurate reports, necessary to assess the safety of the drug for use in clinical investigation; and

(B) adequate information on the chemistry and manufacturing of the drug, controls available for the drug, and primary data tabulations from animal or human studies.

(3)(A) At any time, the Secretary may prohibit the sponsor of an investigation from conducting the investigation (referred to in this paragraph as a ``clinical hold'') if the Secretary makes a determination described in subparagraph (B). The Secretary shall specify the basis for the clinical hold, including the specific information available to the Secretary which served as the basis for such clinical hold, and confirm such determination in writing.

(B) For purposes of subparagraph (A), a determination described in this subparagraph with respect to a clinical hold is that--

(i) the drug involved represents an unreasonable risk to the safety of the persons who are the subjects of the clinical investigation, taking into account the qualifications of the clinical investigators, information about the drug, the design of the clinical investigation, the condition for which the drug is to be investigated, and the health status of the subjects involved; or

(ii) the clinical hold should be issued for such other reasons as the Secretary may by regulation establish (including reasons established by regulation before November 21, 1997).

(C) Any written request to the Secretary from the sponsor of an investigation that a clinical hold be removed shall receive a decision, in writing and specifying the reasons therefor, within 30 days after receipt of such request. Any such request shall include sufficient information to support the removal of such clinical hold.

(4) Regulations under paragraph (1) shall provide that such exemption shall be conditioned upon the manufacturer, or the sponsor of the investigation, requiring that experts using such drugs for investigational purposes certify to such manufacturer or sponsor that they will inform any human beings to whom such drugs, or any controls used in connection therewith, are being administered, or their representatives, that such drugs are being used for investigational purposes and will obtain the consent of such human beings or their representatives, except where it is not feasible or it is contrary to the best interests of such human beings. Nothing in this subsection shall be construed to require any clinical investigator to submit directly to the Secretary reports on the investigational use of drugs.

(j) Abbreviated new drug applications

(1) Any person may file with the Secretary an abbreviated application for the approval of a new drug.

(2)(A) An abbreviated application for a new drug shall contain--

(i) information to show that the conditions of use prescribed, recommended, or suggested in the labeling proposed for the new drug have been previously approved for a drug listed under paragraph (7) (hereinafter in this subsection referred to as a ``listed drug'');

(ii)(I) if the listed drug referred to in clause (i) has only one active ingredient, information to show that the active ingredient of the new drug is the same as that of the listed drug;

(II) if the listed drug referred to in clause (i) has more than one active ingredient, information to show that the active ingredients of the new drug are the same as those of the listed drug, or

(III) if the listed drug referred to in clause (i) has more than one active ingredient and if one of the active ingredients of the new drug is different and the application is filed pursuant to the approval of a petition filed under subparagraph (C), information to show that the other active ingredients of the new drug are the same as the active ingredients of the listed drug, information to show that the different active ingredient is an active ingredient of a listed drug or of a drug which does not meet the requirements of section 321(p) of this title, and such other information respecting the different active ingredient with respect to which the petition was filed as the Secretary may require;

(iii) information to show that the route of administration, the dosage form, and the strength of the new drug are the same as those of the listed drug referred to in clause (i) or, if the route of administration, the dosage form, or the strength of the new drug is different and the application is filed pursuant to the approval of a petition filed under subparagraph (C), such information respecting the route of administration, dosage form, or strength with respect to which the petition was filed as the Secretary may require;

(iv) information to show that the new drug is bioequivalent to the listed drug referred to in clause (i), except that if the application is filed pursuant to the approval of a petition filed under subparagraph (C), information to show that the active ingredients of the new drug are of the same pharmacological or therapeutic class as those of the listed drug referred to in clause (i) and the new drug can be expected to have the same therapeutic effect as the listed drug when administered to patients for a condition of use referred to in clause (i);

(v) information to show that the labeling proposed for the new drug is the same as the labeling approved for the listed drug referred to in clause (i) except for changes required because of differences approved under a petition filed under subparagraph (C) or because the new drug and the listed drug are produced or distributed by different manufacturers;

(vi) the items specified in clauses (B) through (F) of subsection (b)(1) of this section;

(vii) a certification, in the opinion of the applicant and to the best of his knowledge, with respect to each patent which claims the listed drug referred to in clause (i) or which claims a use for such listed drug for which the applicant is seeking approval under this subsection and for which information is required to be filed under subsection (b) or (c) of this section--

(I) that such patent information has not been filed,

(II) that such patent has expired,

(III) of the date on which such patent will expire, or

(IV) that such patent is invalid or will not be infringed by the manufacture, use, or sale of the new drug for which the application is submitted; and

(viii) if with respect to the listed drug referred to in clause (i) information was filed under subsection (b) or (c) of this section for a method of use patent which does not claim a use for which the applicant is seeking approval under this subsection, a statement that the method of use patent does not claim such a use.

The Secretary may not require that an abbreviated application contain information in addition to that required by clauses (i) through (viii).

(B)(i) An applicant who makes a certification described in subparagraph (A)(vii)(IV) shall include in the application a statement that the applicant will give the notice required by clause (ii) to--

(I) each owner of the patent which is the subject of the certification or the representative of such owner designated to receive such notice, and

(II) the holder of the approved application under subsection (b) of this section for the drug which is claimed by the patent or a use of which is claimed by the patent or the representative of such holder designated to receive such notice.

(ii) The notice referred to in clause (i) shall state that an application, which contains data from bioavailability or bioequivalence studies, has been submitted under this subsection for the drug with respect to which the certification is made to obtain approval to engage in the commercial manufacture, use, or sale of such drug before the expiration of the patent referred to in the certification. Such notice shall include a detailed statement of the factual and legal basis of the applicant's opinion that the patent is not valid or will not be infringed.

(iii) If an application is amended to include a certification described in subparagraph (A)(vii)(IV), the notice required by clause (ii) shall be given when the amended application is submitted.

(C) If a person wants to submit an abbreviated application for a new drug which has a different active ingredient or whose route of administration, dosage form, or strength differ from that of a listed drug, such person shall submit a petition to the Secretary seeking permission to file such an application. The Secretary shall approve or disapprove a petition submitted under this subparagraph within ninety days of the date the petition is submitted. The Secretary shall approve such a petition unless the Secretary finds--

(i) that investigations must be conducted to show the safety and effectiveness of the drug or of any of its active ingredients, the route of administration, the dosage form, or strength which differ from the listed drug; or

(ii) that any drug with a different active ingredient may not be adequately evaluated for approval as safe and effective on the basis of the information required to be submitted in an abbreviated application.

(3)(A) The Secretary shall issue guidance for the individuals who review applications submitted under paragraph (1), which shall relate to promptness in conducting the review, technical excellence, lack of bias and conflict of interest, and knowledge of regulatory and scientific standards, and which shall apply equally to all individuals who review such applications.

(B) The Secretary shall meet with a sponsor of an investigation or an applicant for approval for a drug under this subsection if the sponsor or applicant makes a reasonable written request for a meeting for the purpose of reaching agreement on the design and size of bioavailability and bioequivalence studies needed for approval of such application. The sponsor or applicant shall provide information necessary for discussion and agreement on the design and size of such studies. Minutes of any such meeting shall be prepared by the Secretary and made available to the sponsor or applicant.

(C) Any agreement regarding the parameters of design and size of bioavailability and bioequivalence studies of a drug under this paragraph that is reached between the Secretary and a sponsor or applicant shall be reduced to writing and made part of the administrative record by the Secretary. Such agreement shall not be changed after the testing begins, except--

(i) with the written agreement of the sponsor or applicant; or

(ii) pursuant to a decision, made in accordance with

subparagraph (D) by the director of the reviewing division, that a substantial scientific issue essential to determining the safety or effectiveness of the drug has been identified after the testing has begun.

(D) A decision under subparagraph (C)(ii) by the director shall be in writing and the Secretary shall provide to the sponsor or applicant an opportunity for a meeting at which the director and the sponsor or applicant will be present and at which the director will document the scientific issue involved.

(E) The written decisions of the reviewing division shall be binding upon, and may not directly or indirectly be changed by, the field or compliance office personnel unless such field or compliance office personnel demonstrate to the reviewing division why such decision should be modified.

(F) No action by the reviewing division may be delayed because of the unavailability of information from or action by field personnel unless the reviewing division determines that a delay is necessary to assure the marketing of a safe and effective drug.

(G) For purposes of this paragraph, the reviewing division is the division responsible for the review of an application for approval of a drug under this subsection (including scientific matters, chemistry, manufacturing, and controls).

(4) Subject to paragraph (5), the Secretary shall approve an application for a drug unless the Secretary finds--

(A) the methods used in, or the facilities and controls used for, the manufacture, processing, and packing of the drug are inadequate to assure and preserve its identity, strength, quality, and purity;

(B) information submitted with the application is insufficient to show that each of the proposed conditions of use have been previously approved for the listed drug referred to in the application;

(C)(i) if the listed drug has only one active ingredient, information submitted with the application is insufficient to show that the active ingredient is the same as that of the listed drug;

(ii) if the listed drug has more than one active ingredient, information submitted with the application is insufficient to show that the active ingredients are the same as the active ingredients of the listed drug, or

(iii) if the listed drug has more than one active ingredient and if the application is for a drug which has an active ingredient different from the listed drug, information submitted with the application is insufficient to show--

(I) that the other active ingredients are the same as the active ingredients of the listed drug, or

(II) that the different active ingredient is an active ingredient of a listed drug or a drug which does not meet the requirements of section 321(p) of this title,

or no petition to file an application for the drug with the different ingredient was approved under paragraph (2)(C);

(D)(i) if the application is for a drug whose route of administration, dosage form, or strength of the drug is the same as the route of administration, dosage form, or strength of the listed drug referred to in the application, information submitted in the application is insufficient to show that the route of administration, dosage form, or strength is the same as that of the listed drug, or

(ii) if the application is for a drug whose route of

administration, dosage form, or strength of the drug is different from that of the listed drug referred to in the application, no petition to file an application for the drug with the different route of administration, dosage form, or strength was approved under paragraph (2)(C);

(E) if the application was filed pursuant to the approval of a petition under paragraph (2)(C), the application did not contain the information required by the Secretary respecting the active ingredient, route of administration, dosage form, or strength which is not the same;

(F) information submitted in the application is insufficient to show that the drug is bioequivalent to the listed drug referred to in the application or, if the application was filed pursuant to a petition approved under paragraph (2)(C), information submitted in the application is insufficient to show that the active ingredients of the new drug are of the same pharmacological or therapeutic class as those of the listed drug referred to in paragraph (2)(A)(i) and that the new drug can be expected to have the same therapeutic effect as the listed drug when administered to patients for a condition of use referred to in such paragraph;

(G) information submitted in the application is insufficient to show that the labeling proposed for the drug is the same as the labeling approved for the listed drug referred to in the application except for changes required because of differences approved under a petition filed under paragraph (2)(C) or because the drug and the listed drug are produced or distributed by different manufacturers;

(H) information submitted in the application or any other information available to the Secretary shows that (i) the inactive ingredients of the drug are unsafe for use under the conditions prescribed, recommended, or suggested in the labeling proposed for the drug, or (ii) the composition of the drug is unsafe under such conditions because of the type or quantity of inactive ingredients included or the manner in which the inactive ingredients are included;

(I) the approval under subsection (c) of this section of the listed drug referred to in the application under this subsection has been withdrawn or suspended for grounds described in the first sentence of subsection (e) of this section, the Secretary has published a notice of opportunity for hearing to withdraw approval of the listed drug under subsection (c) of this section for grounds described in the first sentence of subsection (e) of this section, the approval under this subsection of the listed drug referred to in the application under this subsection has been withdrawn or suspended under paragraph (6), or the Secretary has determined that the listed drug has been withdrawn from sale for safety or effectiveness reasons;

(J) the application does not meet any other requirement of paragraph (2)(A); or

(K) the application contains an untrue statement of material fact.

(5)(A) Within one hundred and eighty days of the initial receipt of an application under paragraph (2) or within such additional period as may be agreed upon by the Secretary and the applicant, the Secretary shall approve or disapprove the application.

(B) The approval of an application submitted under paragraph (2) shall be made effective on the last applicable date determined under the following:

(i) If the applicant only made a certification described in subclause (I) or (II) of paragraph (2)(A)(vii) or in both such

subclauses, the approval may be made effective immediately.

(ii) If the applicant made a certification described in subclause (III) of paragraph (2)(A)(vii), the approval may be made effective on the date certified under subclause (III).

(iii) If the applicant made a certification described in subclause (IV) of paragraph (2)(A)(vii), the approval shall be made effective immediately unless an action is brought for infringement of a patent which is the subject of the certification before the expiration of forty-five days from the date the notice provided under paragraph (2)(B)(i) is received. If such an action is brought before the expiration of such days, the approval shall be made effective upon the expiration of the thirty-month period beginning on the date of the receipt of the notice provided under paragraph (2)(B)(i) or such shorter or longer period as the court may order because either party to the action failed to reasonably cooperate in expediting the action, except that--

(I) if before the expiration of such period the court decides that such patent is invalid or not infringed, the approval shall be made effective on the date of the court decision,

(II) if before the expiration of such period the court decides that such patent has been infringed, the approval shall be made effective on such date as the court orders under section 271(e)(4)(A) of title 35, or

(III) if before the expiration of such period the court grants a preliminary injunction prohibiting the applicant from engaging in the commercial manufacture or sale of the drug until the court decides the issues of patent validity and infringement and if the court decides that such patent is invalid or not infringed, the approval shall be made effective on the date of such court decision.

In such an action, each of the parties shall reasonably cooperate in expediting the action. Until the expiration of forty-five days from the date the notice made under paragraph (2)(B)(i) is received, no action may be brought under section 2201 of title 28, for a declaratory judgment with respect to the patent. Any action brought under section 2201 shall be brought in the judicial district where the defendant has its principal place of business or a regular and established place of business.

(iv) If the application contains a certification described in subclause (IV) of paragraph (2)(A)(vii) and is for a drug for which a previous application has been submitted under this subsection continuing such a certification, the application shall be made effective not earlier than one hundred and eighty days after--

(I) the date the Secretary receives notice from the applicant under the previous application of the first commercial marketing of the drug under the previous application, or

(II) the date of a decision of a court in an action described in clause (iii) holding the patent which is the subject of the certification to be invalid or not infringed,

whichever is earlier.

(C) If the Secretary decides to disapprove an application, the Secretary shall give the applicant notice of an opportunity for a hearing before the Secretary on the question of whether such application is approvable. If the applicant elects to accept the opportunity for hearing by written request within thirty days after such notice, such hearing shall commence not more than ninety days after the expiration of

such thirty days unless the Secretary and the applicant otherwise agree. Any such hearing shall thereafter be conducted on an expedited basis and the Secretary's order thereon shall be issued within ninety days after the date fixed by the Secretary for filing final briefs.

(D)(i) If an application (other than an abbreviated new drug application) submitted under subsection (b) of this section for a drug, no active ingredient (including any ester or salt of the active ingredient) of which has been approved in any other application under subsection (b) of this section, was approved during the period beginning January 1, 1982, and ending on September 24, 1984, the Secretary may not make the approval of an application submitted under this subsection which refers to the drug for which the subsection (b) application was submitted effective before the expiration of ten years from the date of the approval of the application under subsection (b) of this section.

(ii) If an application submitted under subsection (b) of this section for a drug, no active ingredient (including any ester or salt of the active ingredient) of which has been approved in any other application under subsection (b) of this section, is approved after September 24, 1984, no application may be submitted under this subsection which refers to the drug for which the subsection (b) application was submitted before the expiration of five years from the date of the approval of the application under subsection (b) of this section, except that such an application may be submitted under this subsection after the expiration of four years from the date of the approval of the subsection (b) application if it contains a certification of patent invalidity or noninfringement described in subclause (IV) of paragraph (2)(A)(vii). The approval of such an application shall be made effective in accordance with subparagraph (B) except that, if an action for patent infringement is commenced during the one-year period beginning forty-eight months after the date of the approval of the subsection (b) application, the thirty-month period referred to in subparagraph (B)(iii) shall be extended by such amount of time (if any) which is required for seven and one-half years to have elapsed from the date of approval of the subsection (b) application.

(iii) If an application submitted under subsection (b) of this section for a drug, which includes an active ingredient (including any ester or salt of the active ingredient) that has been approved in another application approved under subsection (b) of this section, is approved after September 24, 1984, and if such application contains reports of new clinical investigations (other than bioavailability studies) essential to the approval of the application and conducted or sponsored by the applicant, the Secretary may not make the approval of an application submitted under this subsection for the conditions of approval of such drug in the subsection (b) application effective before the expiration of three years from the date of the approval of the application under subsection (b) of this section for such drug.

(iv) If a supplement to an application approved under subsection (b) of this section is approved after September 24, 1984, and the supplement contains reports of new clinical investigations (other than bioavailability studies) essential to the approval of the supplement and conducted or sponsored by the person submitting the supplement, the Secretary may not make the approval of an application submitted under this subsection for a change approved in the supplement effective before the expiration of three years from the date of the approval of the supplement under subsection (b) of this section.

(v) If an application (or supplement to an application) submitted under subsection (b) of this section for a drug, which includes an active ingredient (including any ester or salt of the active ingredient) that has been approved in another application under subsection (b) of this section, was approved during the period beginning January 1, 1982,

and ending on September 24, 1984, the Secretary may not make the approval of an application submitted under this subsection which refers to the drug for which the subsection (b) application was submitted or which refers to a change approved in a supplement to the subsection (b) application effective before the expiration of two years from September 24, 1984.

(6) If a drug approved under this subsection refers in its approved application to a drug the approval of which was withdrawn or suspended for grounds described in the first sentence of subsection (e) of this section or was withdrawn or suspended under this paragraph or which, as determined by the Secretary, has been withdrawn from sale for safety or effectiveness reasons, the approval of the drug under this subsection shall be withdrawn or suspended--

(A) for the same period as the withdrawal or suspension under subsection (e) of this section or this paragraph, or

(B) if the listed drug has been withdrawn from sale, for the period of withdrawal from sale or, if earlier, the period ending on the date the Secretary determines that the withdrawal from sale is not for safety or effectiveness reasons.

(7)(A)(i) Within sixty days of September 24, 1984, the Secretary shall publish and make available to the public--

(I) a list in alphabetical order of the official and proprietary name of each drug which has been approved for safety and effectiveness under subsection (c) of this section before September 24, 1984;

(II) the date of approval if the drug is approved after 1981 and the number of the application which was approved; and

(III) whether in vitro or in vivo bioequivalence studies, or both such studies, are required for applications filed under this subsection which will refer to the drug published.

(ii) Every thirty days after the publication of the first list under clause (i) the Secretary shall revise the list to include each drug which has been approved for safety and effectiveness under subsection (c) of this section or approved under this subsection during the thirty-day period.

(iii) When patent information submitted under subsection (b) or (c) of this section respecting a drug included on the list is to be published by the Secretary, the Secretary shall, in revisions made under clause (ii), include such information for such drug.

(B) A drug approved for safety and effectiveness under subsection (c) of this section or approved under this subsection shall, for purposes of this subsection, be considered to have been published under subparagraph (A) on the date of its approval or September 24, 1984, whichever is later.

(C) If the approval of a drug was withdrawn or suspended for grounds described in the first sentence of subsection (e) of this section or was withdrawn or suspended under paragraph (6) or if the Secretary determines that a drug has been withdrawn from sale for safety or effectiveness reasons, it may not be published in the list under subparagraph (A) or, if the withdrawal or suspension occurred after its publication in such list, it shall be immediately removed from such list--

(i) for the same period as the withdrawal or suspension under subsection (e) of this section or paragraph (6), or

(ii) if the listed drug has been withdrawn from sale, for the period of withdrawal from sale or, if earlier, the period ending on the date the Secretary determines that the withdrawal from sale is not for safety or effectiveness reasons.

A notice of the removal shall be published in the Federal Register.

(8) For purposes of this subsection:

(A) The term ``bioavailability'' means the rate and extent to which the active ingredient or therapeutic ingredient is absorbed from a drug and becomes available at the site of drug action.

(B) A drug shall be considered to be bioequivalent to a listed drug if--

(i) the rate and extent of absorption of the drug do not show a significant difference from the rate and extent of absorption of the listed drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses; or

(ii) the extent of absorption of the drug does not show a significant difference from the extent of absorption of the listed drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses and the difference from the listed drug in the rate of absorption of the drug is intentional, is reflected in its proposed labeling, is not essential to the attainment of effective body drug concentrations on chronic use, and is considered medically insignificant for the drug.

(9) The Secretary shall, with respect to each application submitted under this subsection, maintain a record of--

(A) the name of the applicant,

(B) the name of the drug covered by the application,

(C) the name of each person to whom the review of the chemistry of the application was assigned and the date of such assignment, and

(D) the name of each person to whom the bioequivalence review for such application was assigned and the date of such assignment.

The information the Secretary is required to maintain under this paragraph with respect to an application submitted under this subsection shall be made available to the public after the approval of such application.

(k) Records and reports; required information; regulations and orders; access to records

(1) In the case of any drug for which an approval of an application filed under subsection (b) or (j) of this section is in effect, the applicant shall establish and maintain such records, and make such reports to the Secretary, of data relating to clinical experience and other data or information, received or otherwise obtained by such applicant with respect to such drug, as the Secretary may by general regulation, or by order with respect to such application, prescribe on the basis of a finding that such records and reports are necessary in order to enable the Secretary to determine, or facilitate a determination, whether there is or may be ground for invoking subsection (e) of this section. Regulations and orders issued under this subsection and under subsection (i) of this section shall have due regard for the professional ethics of the medical profession and the interests of patients and shall provide, where the Secretary deems it to be appropriate, for the examination, upon request, by the persons to whom such regulations or orders are applicable, of similar information received or otherwise obtained by the Secretary.

(2) Every person required under this section to maintain records,

and every person in charge or custody thereof, shall, upon request of an officer or employee designated by the Secretary, permit such officer or employee at all reasonable times to have access to and copy and verify such records.

(l) Public disclosure of safety and effectiveness data

Safety and effectiveness data and information which has been submitted in an application under subsection (b) of this section for a drug and which has not previously been disclosed to the public shall be made available to the public, upon request, unless extraordinary circumstances are shown--

(1) if no work is being or will be undertaken to have the application approved,

(2) if the Secretary has determined that the application is not approvable and all legal appeals have been exhausted,

(3) if approval of the application under subsection (c) of this section is withdrawn and all legal appeals have been exhausted,

(4) if the Secretary has determined that such drug is not a new drug, or

(5) upon the effective date of the approval of the first application under subsection (j) of this section which refers to such drug or upon the date upon which the approval of an application under subsection (j) of this section which refers to such drug could be made effective if such an application had been submitted.

(m) ``Patent'' defined

For purposes of this section, the term ``patent'' means a patent issued by the United States Patent and Trademark Office.

(n) Scientific advisory panels

(1) For the purpose of providing expert scientific advice and recommendations to the Secretary regarding a clinical investigation of a drug or the approval for marketing of a drug under this section or section 262 of title 42, the Secretary shall establish panels of experts or use panels of experts established before November 21, 1997, or both.

(2) The Secretary may delegate the appointment and oversight authority granted under section 394 of this title to a director of a center or successor entity within the Food and Drug Administration.

(3) The Secretary shall make appointments to each panel established under paragraph (1) so that each panel shall consist of--

(A) members who are qualified by training and experience to evaluate the safety and effectiveness of the drugs to be referred to the panel and who, to the extent feasible, possess skill and experience in the development, manufacture, or utilization of such drugs;

(B) members with diverse expertise in such fields as clinical and administrative medicine, pharmacy, pharmacology, pharmacoeconomics, biological and physical sciences, and other related professions;

(C) a representative of consumer interests, and a representative of interests of the drug manufacturing industry not directly affected by the matter to be brought before the panel; and

(D) two or more members who are specialists or have other expertise in the particular disease or condition for which the drug under review is proposed to be indicated.

Scientific, trade, and consumer organizations shall be afforded an

opportunity to nominate individuals for appointment to the panels. No individual who is in the regular full-time employ of the United States and engaged in the administration of this chapter may be a voting member of any panel. The Secretary shall designate one of the members of each panel to serve as chairman thereof.

(4) Each member of a panel shall publicly disclose all conflicts of interest that member may have with the work to be undertaken by the panel. No member of a panel may vote on any matter where the member or the immediate family of such member could gain financially from the advice given to the Secretary. The Secretary may grant a waiver of any conflict of interest requirement upon public disclosure of such conflict of interest if such waiver is necessary to afford the panel essential expertise, except that the Secretary may not grant a waiver for a member of a panel when the member's own scientific work is involved.

(5) The Secretary shall, as appropriate, provide education and training to each new panel member before such member participates in a panel's activities, including education regarding requirements under this chapter and related regulations of the Secretary, and the administrative processes and procedures related to panel meetings.

(6) Panel members (other than officers or employees of the United States), while attending meetings or conferences of a panel or otherwise engaged in its business, shall be entitled to receive compensation for each day so engaged, including traveltime, at rates to be fixed by the Secretary, but not to exceed the daily equivalent of the rate in effect for positions classified above grade GS-15 of the General Schedule. While serving away from their homes or regular places of business, panel members may be allowed travel expenses (including per diem in lieu of subsistence) as authorized by section 5703 of title 5, for persons in the Government service employed intermittently.

(7) The Secretary shall ensure that scientific advisory panels meet regularly and at appropriate intervals so that any matter to be reviewed by such a panel can be presented to the panel not more than 60 days after the matter is ready for such review. Meetings of the panel may be held using electronic communication to convene the meetings.

(8) Within 90 days after a scientific advisory panel makes recommendations on any matter under its review, the Food and Drug Administration official responsible for the matter shall review the conclusions and recommendations of the panel, and notify the affected persons of the final decision on the matter, or of the reasons that no such decision has been reached. Each such final decision shall be documented including the rationale for the decision.

(June 25, 1938, ch. 675, Sec. 505, 52 Stat. 1052; Pub. L. 86-507, Sec. 1(18), June 11, 1960, 74 Stat. 201; Pub. L. 87-781, title I, Secs. 102(b)-(d), 103(a), (b), 104(a)-(d)(2), Oct. 10, 1962, 76 Stat. 781-783, 784, 785; Pub. L. 92-387, Sec. 4(d), Aug. 16, 1972, 86 Stat. 562; Pub. L. 98-417, title I, Secs. 101, 102(a)-(b)(5), 103, 104, Sept. 24, 1984, 98 Stat. 1585, 1592, 1593, 1597; Pub. L. 102-282, Sec. 5, May 13, 1992, 106 Stat. 161; Pub. L. 103-80, Sec. 3(n), Aug. 13, 1993, 107 Stat. 777; Pub. L. 105-115, title I, Secs. 115, 117, 119, 120, 124(a), Nov. 21, 1997, 111 Stat. 2313, 2315, 2316, 2318, 2324; Pub. L. 106-113, div. B, Sec. 1000(a)(9) [title IV, Sec. 4732(b)(11)], Nov. 29, 1999, 113 Stat. 1536, 1501A-584; Pub. L. 107-109, Sec. 15(c)(1), Jan. 4, 2002, 115 Stat. 1420.)

References in Text

The General Schedule, referred to in subsec. (n)(6), is set out under section 5332 of Title 5, Government Organization and Employees.

Amendments

2002--Subsec. (i)(1)(D). Pub. L. 107-109 added subpar. (D).

1999--Subsec. (m). Pub. L. 106-113 substituted ``United States Patent and Trademark Office'' for ``Patent and Trademark Office of the Department of Commerce''.

1997--Subsec. (b)(1). Pub. L. 105-115, Sec. 115(b), inserted at end ``The Secretary shall, in consultation with the Director of the National Institutes of Health and with representatives of the drug manufacturing industry, review and develop guidance, as appropriate, on the inclusion of women and minorities in clinical trials required by clause (A).''

Subsec. (b)(4). Pub. L. 105-115, Sec. 119(a), added par. (4).

Subsec. (c)(4). Pub. L. 105-115, Sec. 124(a), added par. (4).

Subsec. (d). Pub. L. 105-115, Sec. 115(a), inserted at end ``If the Secretary determines, based on relevant science, that data from one adequate and well-controlled clinical investigation and confirmatory evidence (obtained prior to or after such investigation) are sufficient to establish effectiveness, the Secretary may consider such data and evidence to constitute substantial evidence for purposes of the preceding sentence.''

Subsec. (i). Pub. L. 105-115, Sec. 117, inserted ``(1)'' after ``(i)'', redesignated former pars. (1) to (3) as subpars. (A) to (C), respectively, of par. (1), added pars. (2) to (4), and struck out closing provisions which read as follows: ``Such regulations shall provide that such exemption shall be conditioned upon the manufacturer, or the sponsor of the investigation, requiring that experts using such drugs for investigational purposes certify to such manufacturer or sponsor that they will inform any human beings to whom such drugs, or any controls used in connection therewith, are being administered, or their representatives, that such drugs are being used for investigational purposes and will obtain the consent of such human beings or their representatives, except where they deem it not feasible or, in their professional judgment, contrary to the best interests of such human beings. Nothing in this subsection shall be construed to require any clinical investigator to submit directly to the Secretary reports on the investigational use of drugs.''

Subsec. (j)(2)(A)(i). Pub. L. 105-115, Sec. 119(b)(2)(A), substituted ``paragraph (7)'' for ``paragraph (6)''.

Subsec. (j)(3). Pub. L. 105-115, Sec. 119(b)(1)(B), added par. (3). Former par. (3) redesignated (4).

Subsec. (j)(4). Pub. L. 105-115, Sec. 119(b)(1)(A), (2)(B), redesignated par. (3) as (4) and in introductory provisions substituted ``paragraph (5)'' for ``paragraph (4)''. Former par. (4) redesignated (5).

Subsec. (j)(4)(I). Pub. L. 105-115, Sec. 119(b)(2)(C), substituted ``paragraph (6)'' for ``paragraph (5)''.

Subsec. (j)(5), (6). Pub. L. 105-115, Sec. 119(b)(1)(A), redesignated pars. (4) and (5) as (5) and (6), respectively. Former par. (6) redesignated (7).

Subsec. (j)(7). Pub. L. 105-115, Sec. 119(b)(1)(A), (2)(D), redesignated par. (6) as (7) and in subpar. (C) substituted ``paragraph (6)'' for ``paragraph (5)'' in two places. Former par. (7) redesignated (8).

Subsec. (j)(8), (9). Pub. L. 105-115, Sec. 119(b)(1)(A), redesignated pars. (7) and (8) as (8) and (9), respectively.

Subsec. (n). Pub. L. 105-115, Sec. 120, added subsec. (n).

1993--Subsec. (j)(6)(A)(ii). Pub. L. 103-80, Sec. 3(n)(1)(A), substituted ``Secretary'' for ``Secretry''.

Subsec. (j)(6)(A)(iii). Pub. L. 103-80, Sec. 3(n)(1)(B), inserted

comma after ``published by the Secretary''.

Subsec. (k)(1). Pub. L. 103-80, Sec. 3(n)(2), substituted ``section. Regulations'' for ``section: Provided, however, That regulations''.

1992--Subsec. (j)(8). Pub. L. 102-282 added par. (8).

1984--Subsec. (a). Pub. L. 98-417, Sec. 102(b)(1), inserted ``or (j)'' after ``subsection (b)''.

Subsec. (b). Pub. L. 98-417, Secs. 102(a)(1), 103(a), designated existing provisions of subsec. (b) as par. (1) thereof and redesignated existing cls. (1) through (6) of such par. (1) as cls. (A) through (F) thereof, respectively, inserted requirement that the applicant file with the application the patent number and the expiration date of any patent which claims the drug for which the applicant submitted the application or which claims a method of using such drug and with respect to which a claim of patent infringement could reasonably by asserted if a person not licensed by the owner engaged in the manufacture, use, or sale of the drug, that the applicant amend the application to include such information if an application is filed under this subsection for a drug and a patent which claims such drug or a method of using such drug is issued after the filing date but before approval of the application, and that upon approval of the application, the Secretary publish the information submitted, and added pars. (2) and (3).

Subsec. (c). Pub. L. 98-417, Secs. 102(a)(2), (b)(2), 103(b), designated existing provisions of subsec. (c) as par. (1) thereof and in par. (1) as so designated substituted ``subsection (b) of this section'' for ``this subsection'' and redesignated former pars. (1) and (2) as subpars. (A) and (B), respectively, and added pars. (2) and (3).

Subsec. (d)(6), (7). Pub. L. 98-417, Sec. 102(a)(3)(A), added cl. (6) relating to the failure of the application to contain the patent information prescribed by subsec. (b) of this section, and redesignated former cl. (6) as (7).

Subsec. (e). Pub. L. 98-417, Sec. 102(a)(3)(B), in first sentence, added a new cl. (4) relating to the failure to file the patent information prescribed by subsec. (c) of this section within 30 days after the receipt of written notice from the Secretary specifying the failure to file such information, and redesignated former cl. (4) as (5).

Pub. L. 98-417, Sec. 102(b)(3), (4), in second sentence, inserted in provisions preceding cl. (1) ``submitted under subsection (b) or (j) of this section'' and in cl. (1) substituted ``under subsection (k) of this section or to comply with the notice requirements of section 360(k)(2) of this title'' for ``under subsection (j) of this section or to comply with the notice requirements of section 360(j)(2) of this title''.

Subsecs. (j), (k). Pub. L. 98-417, Sec. 101, added subsec. (j) and redesignated former subsec. (j) as (k).

Subsec. (k)(1). Pub. L. 98-417, Sec. 102(b)(5), substituted ``under subsection (b) or (j) of this section'' for ``pursuant to this section''.

Subsecs. (l), (m). Pub. L. 98-417, Sec. 104, added subsecs. (l) and (m).

1972--Subsec. (e). Pub. L. 92-387 inserted ``or to comply with the notice requirements of section 360(j)(2) of this title'' in cl. (1) of second sentence relating to the maintenance of records.

1962--Subsec. (a). Pub. L. 87-781, Sec. 104(a), inserted ``an approval of'' before ``an application''.

Subsec. (b). Pub. L. 87-781, Sec. 102(b), inserted ``and whether such drug is effective in use'' after ``is safe for use''.

Subsec. (c). Pub. L. 87-781, Sec. 104(b), substituted provisions requiring the Secretary, within 180 days after filing an application, or such additional period as the Secretary and the applicant agree upon, to either approve the application, if meeting the requirements of subsec.

(d) of this section, or give notice of opportunity for hearing on question of whether such application is approvable, and providing that if applicant requests hearing in writing within 30 days, the hearing shall begin within 90 days after expiration of said 30 days, unless the Secretary and applicant agree otherwise, that such hearing shall be expedited, and that the Secretary's order shall be issued within 90 days after date for filing final briefs, for provisions which had an application become effective on the sixtieth day after filing thereof unless prior thereto the Secretary postponed the date by written notice to such time, but not more than 180 days after filing, as the Secretary deemed necessary to study and investigate the application.

Subsec. (d). Pub. L. 87-781, Sec. 102(c), inserted references to subsec. (c), added cls. (5) and (6), provided that if after notice and opportunity for hearing, the Secretary finds that cls. (1) to (6) do not apply, he shall approve the application, and defined ``substantial evidence'' as used in this subsection and subsec. (e) of this section.

Subsec. (e). Pub. L. 87-781, Sec. 102(d), amended subsec. (e) generally, and among other changes, directed the Secretary to withdraw approval of an application if by tests, other scientific data or experience, or new evidence of clinical experience not contained in the application or available at the time of its approval, the drug is shown to be unsafe, or on the basis of new information, there is shown a lack of substantial evidence that the drug has the effect it is represented to have, and provided that if the Secretary, or acting Secretary, finds there is an imminent hazard to the public health, he may suspend approval immediately, notify the applicant, and give him opportunity for an expedited hearing, that the Secretary may withdraw approval if the applicant fails to establish a system for maintaining required records, or has repeatedly or deliberately failed to maintain records and make reports, or has refused access to, or copying or verification of such records, or if the Secretary finds on new evidence that the methods, facilities and controls in the manufacturing, processing, and packing are inadequate to assure and preserve the drugs' identity, strength, quality and purity, and were not made adequate within a reasonable time after receipt of written notice thereof, or finds on new evidence, that the labeling is false or misleading and was not corrected within a reasonable time after receipt of written notice thereof.

Subsec. (f). Pub. L. 87-781, Sec. 104(c), substituted provisions requiring the Secretary to revoke any previous order under subsecs. (d) or (e) of this section refusing, withdrawing, or suspending approval of an application and to approve such application or reinstate such approval, for provisions which required him to revoke an order refusing effectiveness to an application.

Subsec. (h). Pub. L. 87-781, Sec. 104(d)(1), (2), inserted ``as provided in section 2112 of title 28'', and ``except that until the filing of the record the Secretary may modify or set aside his order'', substituted ``or withdrawing approval of an application under this section'' for ``to permit the application to become effective, or suspending the effectiveness of the application'', ``United States court of appeals for the circuit'' for ``district court of the United States within any district'', ``Court of Appeals for the District of Columbia Circuit'' for ``District Court for the District of Columbia'', ``transmitted by the clerk of the court to'' for ``served upon'', and ``by the Supreme Court of the United States upon certiorari or certification as provided in section 1254 of title 28'' for ``as provided in sections 225, 346, and 347 of title 28, as amended, and in section 7, as amended, of the Act entitled `An Act to establish a Court of Appeals for the District of Columbia', approved February 9, 1893'', and eliminated ``upon'' before ``any officer designated'', ``a transcript of'' before ``the record'' and ``and decree'' before ``of the

court affirming''.

Subsec. (i). Pub. L. 87-781, Sec. 103(b), inserted ``the foregoing subsections of'' after ``operation of'', and ``and effectiveness'' after ``safety'', and provided that the regulations may condition exemptions upon the submission of reports of preclinical tests to justify the proposed clinical testing, upon the obtaining by the manufacturer or sponsor of the investigation of a new drug of a signed agreement from each of the investigators that patients to whom the drug is administered will be under his supervision or under investigators responsible to him, and that he will not supply such drug to any other investigator, or to clinics, for administration to human beings, or upon the establishment and maintenance of records and reports of data obtained by the investigational use of such drug, as the Secretary finds will enable him to evaluate the safety and effectiveness of such drug, and provided that the regulations shall condition an exemption upon the manufacturer or sponsor of the investigation requiring that experts using such drugs certify that they will inform humans to whom such drugs or any controls connected therewith are administered, or their representatives, and will obtain the consent of such people where feasible and not contrary to the best interests of such people, and that reports on the investigational use of drugs are not required to be submitted directly to the Secretary.

Subsec. (j). Pub. L. 87-781, Sec. 103(a), added subsec. (j).

1960--Subsec. (g). Pub. L. 86-507 inserted ``or by certified mail'' after ``registered mail''.

Effective Date of 1999 Amendment

Amendment by Pub. L. 106-113 effective 4 months after Nov. 29, 1999, see section 1000(a)(9) [title IV, Sec. 4731] of Pub. L. 106-113, set out as a note under section 1 of Title 35, Patents.

Effective Date of 1997 Amendment

Amendment by Pub. L. 105-115 effective 90 days after Nov. 21, 1997, except as otherwise provided, see section 501 of Pub. L. 105-115, set out as a note under section 321 of this title.

Effective Date of 1984 Amendment

Section 105 of Pub. L. 98-417 provided that:

``(a) The Secretary of Health and Human Services shall promulgate, in accordance with the notice and comment requirements of section 553 of title 5, United States Code, such regulations as may be necessary for the administration of section 505 of the Federal Food, Drug, and Cosmetic Act [this section], as amended by sections 101, 102, and 103 of this Act, within one year of the date of enactment of this Act [Sept. 24, 1984].

``(b) During the period beginning sixty days after the date of the enactment of this Act [Sept. 24, 1984], and ending on the date regulations promulgated under subsection (a) take effect, abbreviated new drug applications may be submitted in accordance with the provisions of section 314.2 of title 21 of the Code of Federal Regulations and shall be considered as suitable for any drug which has been approved for safety and effectiveness under section 505(c) of the Federal Food, Drug, and Cosmetic Act [subsec. (c) of this section] before the date of the enactment of this Act. If any such provision is inconsistent with the requirements of section 505(j) of the Federal Food, Drug, and Cosmetic

Act, the Secretary shall consider the application under the applicable requirements of such section. The Secretary of Health and Human Services may not approve such an abbreviated new drug application which is filed for a drug which is described in sections 505(c)(3)(D) and 505(j)(4)(D) of the Federal Food, Drug, and Cosmetic Act, except in accordance with such section.''

Effective Date of 1972 Amendment

Amendment by Pub. L. 92-387 effective on first day of sixth month beginning after Aug. 16, 1972, see section 5 of Pub. L. 92-387, set out as a note under section 360 of this title.

Effective Date of 1962 Amendment

Amendment by Pub. L. 87-781 effective on first day of seventh calendar month following October 1962, see section 107 of Pub. L. 87-781, set out as a note under section 321 of this title.

Construction of Amendments by Pub. L. 102-282

Amendment by Pub. L. 102-282 not to preclude any other civil, criminal, or administrative remedy provided under Federal or State law, including any private right of action against any person for the same action subject to any action or civil penalty under an amendment made by Pub. L. 102-282, see section 7 of Pub. L. 102-282, set out as a note under section 335a of this title.

Transfer of Functions

For transfer of functions of Federal Security Administrator to Secretary of Health, Education, and Welfare [now Health and Human Services], and of Food and Drug Administration in the Department of Agriculture to Federal Security Agency, see note set out under section 41 of this title.

Report on Patient Access to New Therapeutic Agents for Pediatric Cancer

Pub. L. 107-109, Sec. 15(d), Jan. 4, 2002, 115 Stat. 1421, provided that: ``Not later than January 31, 2003, the Secretary of Health and Human Services, acting through the Commissioner of Food and Drugs and in consultation with the Director of the National Institutes of Health, shall submit to the Committee on Health, Education, Labor, and Pensions of the Senate and the Committee on Energy and Commerce of the House of Representatives a report on patient access to new therapeutic agents for pediatric cancer, including access to single patient use of new therapeutic agents.''

Data Requirements for Drugs and Biologics

Section 118 of Pub. L. 105-115 provided that: ``Within 12 months after the date of enactment of this Act [Nov. 21, 1997], the Secretary of Health and Human Services, acting through the Commissioner of Food and Drugs, shall issue guidance that describes when abbreviated study reports may be submitted, in lieu of full reports, with a new drug

application under section 505(b) of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355(b)) and with a biologics license application under section 351 of the Public Health Service Act (42 U.S.C. 262) for certain types of studies. Such guidance shall describe the kinds of studies for which abbreviated reports are appropriate and the appropriate abbreviated report formats.''

Requirements for Review of Approval Procedures and Current Good Manufacturing Practices for Positron Emission Technology

Section 121(c) of Pub. L. 105-115 provided that:
``(1) Procedures and requirements.--
``(A) In general.--In order to take account of the special characteristics of positron emission tomography drugs and the special techniques and processes required to produce these drugs, not later than 2 years after the date of enactment of this Act [Nov. 21, 1997], the Secretary of Health and Human Services shall establish--
``(i) appropriate procedures for the approval of positron emission tomography drugs pursuant to section 505 of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355); and
``(ii) appropriate current good manufacturing practice requirements for such drugs.
``(B) Considerations and consultation.--In establishing the procedures and requirements required by subparagraph (A), the Secretary of Health and Human Services shall take due account of any relevant differences between not-for-profit institutions that compound the drugs for their patients and commercial manufacturers of the drugs. Prior to establishing the procedures and requirements, the Secretary of Health and Human Services shall consult with patient advocacy groups, professional associations, manufacturers, and physicians and scientists licensed to make or use positron emission tomography drugs.
``(2) Submission of new drug applications and abbreviated new drug applications.--
``(A) In general.--Except as provided in subparagraph (B), the Secretary of Health and Human Services shall not require the submission of new drug applications or abbreviated new drug applications under subsection (b) or (j) of section 505 (21 U.S.C. 355), for compounded positron emission tomography drugs that are not adulterated drugs described in section 501(a)(2)(C) of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 351(a)(2)(C)) (as amended by subsection (b)), for a period of 4 years after the date of enactment of this Act [Nov. 21, 1997], or for 2 years after the date on which the Secretary establishes procedures and requirements under paragraph (1), whichever is longer.
``(B) Exception.--Nothing in this Act [see Short Title of 1997 Amendment note set out under section 301 of this title] shall prohibit the voluntary submission of such applications or the review of such applications by the Secretary of Health and Human Services. Nothing in this Act shall constitute an exemption for a positron emission tomography drug from the requirements of regulations issued under section 505(i) of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355(i)).''

``Compounded Positron Emission Topography Drug'' Defined

Section 121(e) of Pub. L. 105-115 provided that: ``As used in this

section [amending sections 321 and 351 of this title and enacting provisions set out as notes under this section and section 351 of this title], the term `compounded positron emission tomography drug' has the meaning given the term in section 201 of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 321).''

Requirements for Radiopharmaceuticals

Section 122 of Pub. L. 105-115 provided that:

``(a) Requirements.--

``(1) Regulations.--

``(A) Proposed regulations.--Not later than 180 days after the date of enactment of this Act [Nov. 21, 1997], the Secretary of Health and Human Services, after consultation with patient advocacy groups, associations, physicians licensed to use radiopharmaceuticals, and the regulated industry, shall issue proposed regulations governing the approval of radiopharmaceuticals. The regulations shall provide that the determination of the safety and effectiveness of such a radiopharmaceutical under section 505 of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355) or section 351 of the Public Health Service Act (42 U.S.C. 262) shall include consideration of the proposed use of the radiopharmaceutical in the practice of medicine, the pharmacological and toxicological activity of the radiopharmaceutical (including any carrier or ligand component of the radiopharmaceutical), and the estimated absorbed radiation dose of the radiopharmaceutical.

``(B) Final regulations.--Not later than 18 months after the date of enactment of this Act, the Secretary shall promulgate final regulations governing the approval of the radiopharmaceuticals.

``(2) Special rule.--In the case of a radiopharmaceutical, the indications for which such radiopharmaceutical is approved for marketing may, in appropriate cases, refer to manifestations of disease (such as biochemical, physiological, anatomic, or pathological processes) common to, or present in, one or more disease states.

``(b) Definition.--In this section, the term `radiopharmaceutical' means--

``(1) an article--

``(A) that is intended for use in the diagnosis or monitoring of a disease or a manifestation of a disease in humans; and

``(B) that exhibits spontaneous disintegration of unstable nuclei with the emission of nuclear particles or photons; or

``(2) any nonradioactive reagent kit or nuclide generator that is intended to be used in the preparation of any such article.''

Special Rule

Section 123(f) of Pub. L. 105-115 provided that: ``The Secretary of Health and Human Services shall take measures to minimize differences in the review and approval of products required to have approved biologics license applications under section 351 of the Public Health Service Act (42 U.S.C. 262) and products required to have approved new drug applications under section 505(b)(1) of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355(b)(1)).''

Transition

Section 125(d) of Pub. L. 105-115 provided that:

``(1) In general.--An application that was approved by the Secretary of Health and Human Services before the date of the enactment of this Act [Nov. 21, 1997] for the marketing of an antibiotic drug under section 507 of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 357), as in effect on the day before the date of the enactment of this Act, shall, on and after such date of enactment, be considered to be an application that was submitted and filed under section 505(b) of such Act (21 U.S.C. 355(b)) and approved for safety and effectiveness under section 505(c) of such Act (21 U.S.C. 355(c)), except that if such application for marketing was in the form of an abbreviated application, the application shall be considered to have been filed and approved under section 505(j) of such Act (21 U.S.C. 355(j)).

``(2) Exception.--The following subsections of section 505 (21 U.S.C. 355) shall not apply to any application for marketing in which the drug that is the subject of the application contains an antibiotic drug and the antibiotic drug was the subject of any application for marketing received by the Secretary of Health and Human Services under section 507 of such Act (21 U.S.C. 357) before the date of the enactment of this Act [Nov. 21, 1997]:

``(A)(i) Subsections (c)(2), (d)(6), (e)(4), (j)(2)(A)(vii), (j)(2)(A)(viii), (j)(2)(B), (j)(4)(B), and (j)(4)(D); and

``(ii) The third and fourth sentences of subsection (b)(1) (regarding the filing and publication of patent information); and

``(B) Subsections (b)(2)(A), (b)(2)(B), (b)(3), and (c)(3) if the investigations relied upon by the applicant for approval of the application were not conducted by or for the applicant and for which the applicant has not obtained a right of reference or use from the person by or for whom the investigations were conducted.

``(3) Publication.--For purposes of this section, the Secretary is authorized to make available to the public the established name of each antibiotic drug that was the subject of any application for marketing received by the Secretary for Health and Human Services under section 507 of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 357) before the date of enactment of this Act [Nov. 21, 1997].''

Termination of Advisory Panels

Advisory panels established after Jan. 5, 1973, to terminate not later than the expiration of the 2-year period beginning on the date of their establishment, unless, in the case of a panel established by the President or an officer of the Federal Government, such panel is renewed by appropriate action prior to the expiration of such 2-year period, or in the case of a panel established by Congress, its duration is otherwise provided for by law. See sections 3(2) and 14 of Pub. L. 92-463, Oct. 6, 1972, 86 Stat. 770, 776, set out in the Appendix to Title 5, Government Organization and Employees.

Appeals Taken Prior to October 10, 1962

Section 104(d)(3) of Pub. L. 87-781 made amendments to subsec. (h) of this section inapplicable to any appeal taken prior to Oct. 10, 1962.

Section Referred to in Other Sections

This section is referred to in sections 321, 331, 333, 334, 335a, 352, 353, 353a, 355a, 355b, 356, 356a, 356c, 360, 360b, 360j, 360aa to 360ee, 360aaa, 360bbb, 360bbb-1, 374, 379g, 379h, 379r, 381, 382, 384, 802, 811, 827 of this title; title 10 section 1107; title 26 section 45C; title 28 section 2201; title 35 sections 155A, 156, 271; title 42 sections 236, 262, 282, 284m, 300cc-12, 300cc-13, 300cc-17, 1395y, 1396r-8.

(*e*) Quantitative or semiquantitative formulas.

(*f*) Information on product design or construction.

(ii) Material submitted under paragraph (j)(2) of this section is to be segregated from all other submitted material and clearly so marked. A person who does not agree that a submission is properly subject to paragraph (j)(2) may request a ruling from the Associate Commissioner for Public Affairs whose decision is final, subject to judicial review under § 20.48.

(3) Material listed in paragraph (j)(2)(i) (*a*) and (*b*) of this section may be disclosed under a protective order issued by the administrative law judge or other presiding officer at a hearing referenced in paragraph (j)(2)(i). The administrative law judge or presiding officer shall permit disclosure of the data only in camera and only to the extent necessary for the proper conduct of the hearing. The administrative law judge or presiding officer shall direct to whom the information is to be made available (e.g., to parties or participants, or only to counsel for parties or participants), and persons not specifically permitted access to the data will be excluded from the in camera part of the proceeding. The administrative law judge or other presiding officer may impose other conditions or safeguards. The limited availability of material under this paragraph does not constitute prior disclosure to the public as defined in § 20.81, and no information subject to a particular order is to be submitted to or received or considered by FDA in support of a petition or other request from any other person.

[44 FR 22323, Apr. 13, 1979, as amended at 46 FR 8455, Jan. 27, 1981; 49 FR 7363, Feb. 29, 1984; 54 FR 9034, Mar. 3, 1989; 59 FR 14363, Mar. 28, 1994; 64 FR 69190, Dec. 10, 1999; 65 FR 56477, Sept. 19, 2000; 66 FR 56035, Nov. 6, 2001; 66 FR 66742, Dec. 27, 2001; 68 FR 25285, May 12, 2003]

§ 10.25 Initiation of administrative proceedings.

An administrative proceeding may be initiated in the following three ways:

(a) An interested person may petition the Commissioner to issue, amend, or revoke a regulation or order, or to take or refrain from taking any other form of administrative action. A petition must be either: (1) In the form specified in other applicable FDA regulations, e.g., the form for a color additive petition in § 71.1, for a food additive petition in § 171.1, for a new drug application in § 314.50, for a new animal drug application in § 514.1, or (2) in the form for a citizen petition in § 10.30.

(b) The Commissioner may initiate a proceeding to issue, amend, or revoke a regulation or order or take or refrain from taking any other form of administrative action. FDA has primary jurisdiction to make the initial determination on issues within its statutory mandate, and will request a court to dismiss, or to hold in abeyance its determination of or refer to the agency for administrative determination, any issue which has not previously been determined by the agency or which, if it has previously been determined, the agency concluded should be reconsidered and subject to a new administrative determination. The Commissioner may utilize any of the procedures established in this part in reviewing and making a determination on any matter initiated under this paragraph.

(c) The Commissioner will institute a proceeding to determine whether to issue, amend, or revoke a regulation or order, or take or refrain from taking any other form of administrative action whenever any court, on its own initiative, holds in abeyance or refers any matter to the agency for an administrative determination and the Commissioner concludes that an administrative determination is feasible within agency priorities and resources.

[44 FR 22323, Apr. 13, 1979, as amended at 54 FR 9034, Mar. 3, 1989]

§ 10.30 Citizen petition.

(a) This section applies to any petition submitted by a person (including a person who is not a citizen of the United States) except to the extent that other sections of this chapter apply different requirements to a particular matter.

(b) A petition (including any attachments) must be submitted in accordance with § 10.20 and in the following form:

(Date) _____

Division of Dockets Management, Food and Drug Administration, Department of Health and Human Services, 5630 Fishers Lane, rm. 1061, Rockville, MD 20852.

CITIZEN PETITION

The undersigned submits this petition under _____ (relevant statutory sections, if known) of the _____ (Federal Food, Drug, and Cosmetic Act or the Public Health Service Act or any other statutory provision for which authority has been delegated to the Commissioner of Food and Drugs under 21 CFR 5.10) to request the Commissioner of Food and Drugs to _____ (issue, amend, or revoke a regulation or order or take or refrain from taking any other form of administrative action).

A. Action requested

((1) If the petition requests the Commissioner to issue, amend, or revoke a regulation, the exact wording of the existing regulation (if any) and the proposed regulation or amendment requested.)

((2) If the petition requests the Commissioner to issue, amend, or revoke an order, a copy of the exact wording of the citation to the existing order (if any) and the exact wording requested for the proposed order.)

((3) If the petition requests the Commissioner to take or refrain from taking any other form of administrative action, the specific action or relief requested.)

B. Statement of grounds

(A full statement, in a well organized format, of the factual and legal grounds on which the petitioner relies, including all relevant information and views on which the petitioner relies, as well as representative information known to the petitioner which is unfavorable to the petitioner's position.)

C. Environmental impact

(A) Claim for categorical exclusion under §§ 25.30, 25.31, 25.32, 25.33, or § 25.34 of this chapter or an environmental assessment under § 25.40 of this chapter.)

D. Economic impact

(The following information is to be submitted only when requested by the Commissioner following review of the petition: A statement of the effect of requested action on: (1) Cost (and price) increases to industry, government, and consumers; (2) productivity of wage earners, businesses, or government; (3) competition; (4) supplies of important materials, products, or services; (5) employment; and (6) energy supply or demand.)

E. Certification

The undersigned certifies, that, to the best knowledge and belief of the undersigned, this petition includes all information and views on which the petition relies, and that it includes representative data and information known to the petitioner which are unfavorable to the petition.

(Signature) _____

(Name of petitioner) _____

(Mailing address) _____

(Telephone number) _____

(c) A petition which appears to meet the requirements of paragraph (b) of this section and § 10.20 will be filed by the Division of Dockets Management, stamped with the date of filing, and assigned a docket number. The docket number identifies the file established by the Division of Dockets Management for all submissions relating to the petition, as provided in this part. Subsequent submissions relating to the matter must refer to the docket number and will be filed in the docket file. Related petitions may be filed together and given the same docket number. The Division of Dockets Management will promptly notify the petitioner in writing of the filing and docket number of a petition.

(d) An interested person may submit written comments to the Division of Dockets Management on a filed petition, which comments become part of the docket file. The comments are to specify the docket number of the petition and may support or oppose the petition in whole or in part. A request for alternative or different administrative action must be submitted as a separate petition.

(e)(1) The Commissioner shall, in accordance with paragraph (e)(2), rule upon each petition filed under paragraph (c) of this section, taking into consideration (i) available agency resources for the category of subject matter, (ii) the priority assigned to the petition considering both the category of subject matter involved and the overall work of the agency, and (iii) time requirements established by statute.

(2) Except as provided in paragraph (e)(4) of this section, the Commissioner shall furnish a response to each petitioner within 180 days of receipt of the petition. The response will either:

(i) Approve the petition, in which case the Commissioner shall concurrently take appropriate action (e.g.,

publication of a FEDERAL REGISTER notice) implementing the approval;

(ii) Deny the petition; or

(iii) Provide a tentative response, indicating why the agency has been unable to reach a decision on the petition, e.g., because of the existence of other agency priorities, or a need for additional information. The tentative response may also indicate the likely ultimate agency response, and may specify when a final response may be furnished.

(3) The Commissioner may grant or deny such a petition, in whole or in part, and may grant such other relief or take other action as the petition warrants. The petitioner is to be notified in writing of the Commissioner's decision. The decision will be placed in the public docket file in the office of the Division of Dockets Management and may also be in the form of a notice published in the FEDERAL REGISTER.

(4) The Commissioner shall furnish a response to each petitioner within 90 days of receipt of a petition filed under section 505(j)(2)(C) of the act. The response will either approve or disapprove the petition. Agency action on a petition shall be governed by §314.93 of this chapter.

(f) If a petition filed under paragraph (c) of this section requests the Commissioner to issue, amend, or revoke a regulation, §10.40 or §10.50 also apply.

(g) A petitioner may supplement, amend, or withdraw a petition in writing without agency approval and without prejudice to resubmission at anytime until the Commissioner rules on the petition, unless the petition has been referred for a hearing under parts 12, 13, 14, or 15. After a ruling or referral, a petition may be supplemented, amended, or withdrawn only with the approval of the Commissioner. The Commissioner may approve withdrawal, with or without prejudice against resubmission of the petition.

(h) In reviewing a petition the Commissioner may use the following procedures:

(1) Conferences, meetings, discussions, and correspondence under §10.65.

(2) A hearing under parts 12, 13, 14, 15, or 16.

(3) A FEDERAL REGISTER notice requesting information and views.

(4) A proposal to issue, amend, or revoke a regulation, in accordance with §10.40 or §12.20.

(5) Any other specific public procedure established in this chapter and expressly applicable to the matter.

(i) The record of the administrative proceeding consists of the following:

(1) The petition, including all information on which it relies, filed by the Division of Dockets Management.

(2) All comments received on the petition, including all information submitted as a part of the comments.

(3) If the petition resulted in a proposal to issue, amend, or revoke a regulation, all of the documents specified in §10.40(g).

(4) The record, consisting of any transcripts, minutes of meetings, reports, FEDERAL REGISTER notices, and other documents resulting from the optional procedures specified in paragraph (h) of this section, except a transcript of a closed portion of a public advisory committee meeting.

(5) The Commissioner's decision on the petition, including all information identified or filed by the Commissioner with the Division of Dockets Management as part of the record supporting the decision.

(6) All documents filed with the Division of Dockets Management under §10.65(h).

(7) If a petition for reconsideration or for a stay of action is filed under paragraph (j) of this section, the administrative record specified in §10.33(k) or §10.35(h).

(j) The administrative record specified in paragraph (i) of this section is the exclusive record for the Commissioner's decision. The record of the administrative proceeding closes on the date of the Commissioner's decision unless some other date is specified. Thereafter any interested person may submit a petition for reconsideration under §10.33 or a petition for stay of action under §10.35. A person who wishes to rely upon information or views not included in the administrative record shall submit them to the Commissioner with a new petition to modify the decision in accordance with this section.

(k) This section does not apply to the referral of a matter to a United States

attorney for the initiation of court enforcement action and related correspondence, or to requests, suggestions, and recommendations made informally in routine correspondence received by FDA. Routine correspondence does not constitute a petition within the meaning of this section unless it purports to meet the requirements of this section. Action on routine correspondence does not constitute final administrative action subject to judicial review under § 10.45.

(l) The Division of Dockets Management will maintain a chronological list of each petition filed under this section and § 10.85, but not of petitions submitted elsewhere in the agency under § 10.25(a)(1), showing:

(1) The docket number;

(2) The date the petition was filed by the Division of Dockets Management;

(3) The name of the petitioner;

(4) The subject matter involved; and

(5) The disposition of the petition.

[44 FR 22323, Apr. 13, 1979, as amended at 46 FR 8455, Jan. 27, 1981; 50 16656, Apr. 26, 1985; 54 FR 9034, Mar. 3, 1989; 57 FR 17980, Apr. 28, 1992; 59 FR 14364, Mar. 28, 1994; 62 FR 40592, July 29, 1997; 66 FR 6467, Jan. 22, 2001; 66 FR 12848, Mar. 1, 2001]

§ 10.33 Administrative reconsideration of action.

(a) The Commissioner may at any time reconsider a matter, on the Commissioner's own initiative or on the petition of an interested person.

(b) An interested person may request reconsideration of part or all of a decision of the Commissioner on a petition submitted under § 10.25. Each request for reconsideration must be submitted in accordance with § 10.20 and in the following form no later than 30 days after the date of the decision involved. The Commissioner may, for good cause, permit a petition to be filed after 30 days. In the case of a decision published in the FEDERAL REGISTER, the day of publication is the day of decision.

(Date) _____

Division of Dockets Management, Food and Drug Administration, Department of Health and Human Services, rm. 1-23, 5630 Fishers Lane, rm. 1061, Rockville, MD 20852.

PETITION FOR RECONSIDERATION

[Docket No.]

The undersigned submits this petition for reconsideration of the decision of the Commissioner of Food and Drugs in Docket No. ____.

A. Decision involved

(A concise statement of the decision of the Commissioner which the petitioner wishes to have reconsidered.)

B. Action requested

(The decision which the petitioner requests the Commissioner to make upon reconsideration of the matter.)

C. Statement of grounds

(A full statement, in a well-organized format, of the factual and legal grounds upon which the petitioner relies. The grounds must demonstrate that relevant information and views contained in the administrative record were not previously or not adequately considered by the Commissioner.

(No new information or views may be included in a petition for reconsideration.)

(Signature) _____
(Name of petitioner) _____
(Mailing address) _____
(Telephone number) _____

(c) A petition for reconsideration relating to a petition submitted under § 10.25(a)(2) is subject to the requirements of § 10.30 (c) and (d), except that it is filed in the same docket file as the petition to which it relates.

(d) The Commissioner shall promptly review a petition for reconsideration. The Commissioner may grant the petition when the Commissioner determines it is in the public interest and in the interest of justice. The Commissioner shall grant a petition for reconsideration in any proceeding if the Commissioner determines all of the following apply:

(1) The petition demonstrates that relevant information or views contained in the administrative record were not previously or not adequately considered.

(2) The petitioner's position is not frivolous and is being pursued in good faith.

(3) The petitioner has demonstrated sound public policy grounds supporting reconsideration.

(4) Reconsideration is not outweighed by public health or other public interests.

use in the reference listed drug's labeling for which the applicant seeks approval.

(3) If the proposed drug product is a combination product with one different active ingredient, including a different ester or salt, from the reference listed drug, that the different active ingredient has previously been approved in a listed drug or is a drug that does not meet the definition of "new drug" in section 201(b) of the act.

(e) No later than 90 days after the date a petition that is permitted under paragraph (a) of this section is submitted, FDA will approve or disapprove the petition.

(1) FDA will approve a petition properly submited under this section unless it finds that:

(i) Investigations must be conducted to show the safety and effectiveness of the drug product or of any of its active ingredients, its route of administration, dosage form, or strength which differs from the reference listed drug; or

(ii) For a petition that seeks to change an active ingredient, the drug product that is the subject of the petition is not a combination drug; or

(iii) For a combination drug product that is the subject of the petition and has an active ingredient different from the reference listed drug:

(A) The drug product may not be adequately evaluated for approval as safe and effective on the basis of the information required to be submitted under §314.94; or

(B) The petition does not contain information to show that the different active ingredient of the drug product is of the same pharmacological or therapeutic class as the ingredient of the reference listed drug that is to be changed and that the drug product can be expected to have the same therapeutic effect as the reference listed drug when administered to patients for each condition of use in the listed drug's labeling for which the applicant seeks approval; or

(C) The different active ingredient is not an active ingredient in a listed drug or a drug that meets the requirements of section 201(p) of the act; or

(D) The remaining active ingredients are not identical to those of the listed combination drug; or

(iv) Any of the proposed changes from the listed drug would jeopardize the safe or effective use of the product so as to necessitate significant labeling changes to address the newly introduced safety or effectiveness problem; or

(v) FDA has determined that the reference listed drug has been withdrawn from sale for safety or effectiveness reasons under §314.161, or the reference listed drug has been voluntarily withdrawn from sale and the agency has not determined whether the withdrawal is for safety or effectiveness reasons.

(2) For purposes of this paragraph, "investigations must be conducted" means that information derived from animal or clinical studies is necessary to show that the drug product is safe or effective. Such information may be contained in published or unpublished reports.

(3) If FDA approves a petition submitted under this section, the agency's response may describe what additional information, if any, will be required to support an abbreviated new drug application for the drug product. FDA may, at any time during the course of its review of an abbreviated new drug application, request additional information required to evaluate the change approved under the petition.

(f) FDA may withdraw approval of a petition if the agency receives any information demonstrating that the petition no longer satisfies the conditions under paragraph (e) of this section.

§314.94 Content and format of an abbreviated application.

Abbreviated applications are required to be submitted in the form and contain the information required under this section. Three copies of the application are required, an archival copy, a review copy, and a field copy. FDA will maintain guidance documents on the format and content of applications to assist applicants in their preparation.

(a) *Abbreviated new drug applications.* Except as provided in paragraph (b) of this section, the applicant shall submit

a complete archival copy of the abbreviated new drug application that includes the following:

(1) *Application form.* The applicant shall submit a completed and signed application form that contains the information described under § 314.50(a)(1), (a)(3), (a)(4), and (a)(5). The applicant shall state whether the submission is an abbreviated application under this section or a supplement to an abbreviated application under § 314.97.

(2) *Table of contents.* the archival copy of the abbreviated new drug application is required to contain a table of contents that shows the volume number and page number of the contents of the submission.

(3) *Basis for abbreviated new drug application submission.* An abbreviated new drug application must refer to a listed drug. Ordinarily, that listed drug will be the drug product selected by the agency as the reference standard for conducting bioequivalence testing. The application shall contain:

(i) The name of the reference listed drug, including its dosage form and strength. For an abbreviated new drug application based on an approverd petition under §10.30 of this chapter or §314.93, the reference listed drug must be the same as the listed drug approved in the petition.

(ii) A statement as to whether, according to the information published in the list, the reference listed drug is entitled to a period of marketing exclusivity under section 505(j)(4)(D) of the act.

(iii) For an abbreviated new drug application based on an approved petition under § 10.30 of this chapter or § 314.93, a reference to FDA-assigned docket number for the petition and a copy of FDA's correspondence approving the petition.

(4) *Conditions of use.* (i) A statement that the conditions of use prescribed, recommended, or suggested in the labeling proposed for the drug product have been previously approved for the reference listed drug.

(ii) A reference to the applicant's annotated proposed labeling and to the currently approved labeling for the reference listed drug provided under paragraph (a)(8) of this section.

(5) *Active ingredients.* (i) For a single-active-ingredient drug product, information to show that the active ingredient is the same as that of the reference single-active-ingredient listed drug, as follows:

(A) A statement that the active ingredient of the proposed drug product is the same as that of the reference listed drug.

(B) A reference to the applicant's annotated proposed labeling and to the currently approved labeling for the reference listed drug provided under paragraph (a)(8) of this section.

(ii) For a combination drug product, information to show that the active ingredients are the same as those of the reference listed drug except for any different active ingredient that has been the subject of an approved petition, as follows:

(A) A statement that the active ingredients of the proposed drug product are the same as those of the reference listed drug, or if one of the active ingredients differs from one of the active ingredients of the reference listed drug and the abbreviated application is submitted under the approval of a petition under § 314.93 to vary such active ingredient, information to show that the other active ingredients of the drug product are the same as the other active ingredients of the reference listed drug, information to show that the different active ingredient is an active ingredient of another listed drug or of a drug that does not meet the definition of "new drug" in section 201(p) of the act, and such other information about the different active ingredient that FDA may require.

(B) A reference to the applicant's annotated proposed labeling and to the currently approved labeling for the reference listed drug provided under paragraph (a)(8) of this section.

(6) *Route of administration, dosage form, and strength.* (i) Information to show that the route of administration, dosage form, and strength of the drug product are the same as those of the reference listed drug except for any differences that have been the subject of an approved petition, as follows:

(A) A statement that the route of administration, dosage form, and strength of the proposed drug product

are the same as those of the reference listed drug.

(B) A reference to the applicant's annotated proposed labeling and to the currently approved labeling for the reference listed drug provided under paragraph (a)(8) of this section.

(ii) If the route of administration, dosage form, or strength of the drug product differs from the reference listed drug and the abbreviated application is submitted under an approved petition under §314.93, such information about the different route of administration, dosage form, or strength that FDA may require.

(7) *Bioequivalence.* (i) Information that shows that the drug product is bioequivalent to the reference listed drug upon which the applicant relies; or

(ii) If the abbreviated new drug application is submitted under a petition approved under §314.93, the results of any bioavailability of bioequivalence testing required by the agency, or any other information required by the agency to show that the active ingredients of the proposed drug product are of the same pharmacological or therapeutic class as those in the reference listed drug and that the proposed drug product can be expected to have the same therapeutic effect as the reference listed drug. If the proposed drug product contains a different active ingredient than the reference listed drug, FDA will consider the proposed drug product to have the same therapeutic effect as the reference listed drug if the applicant provides information demonstrating that:

(A) There is an adequate scientific basis for determining that substitution of the specific proposed dose of the different active ingredient for the dose of the member of the same pharmacological or therapeutic class in the reference listed drug will yield a resulting drug product whose safety and effectiveness have not been adversely affected.

(B) The unchanged active ingredients in the proposed drug product are bioequivalent to those in the reference listed drug.

(C) The different active ingredient in the proposed drug product is bioequivalent to an approved dosage form containing that ingredient and approved for the same indication as the proposed drug product or is bioequivalent to a drug product offered for that indication which does not meet the definition of "new drug" under section 201(p) of the act.

(iii) For each in vivo bioequivalence study contained in the abbreviated new drug application, a description of the analytical and statistical methods used in each study and a statement with respect to each study that it either was conducted in compliance with the institutional review board regulations in part 56 of this chapter, or was not subject to the regulations under §56.104 or §56.105 of this chapter and that each study was conducted in compliance with the informed consent regulations in part 50 of this chapter.

(8) *Labeling*—(i) *Listed drug labeling.* A copy of the currently approved labeling (including, if applicable, any Medication Guide required under part 208 of this chapter) for the listed drug referred to in the abbreviated new drug application, if the abbreviated new drug application relies on a reference listed drug.

(ii) *Copies of proposed labeling.* Copies of the label and all labeling for the drug product including, if applicable, any Medication Guide required under part 208 of this chapter (4 copies of draft labeling or 12 copies of final printed labeling).

(iii) *Statement on proposed labeling.* A statement that the applicant's proposed labeling including, if applicable, any Medication Guide required under part 208 of this chapter is the same as the labeling of the reference listed drug except for differences annotated and explained under paragraph (a)(8)(iv) of this section.

(iv) *Comparison of approved and proposed labeling.* A side-by-side comparison of the applicant's proposed labeling including, if applicable, any Medication Guide required under part 208 of this chapter with the approved labeling for the reference listed drug with all differences annotated and explained. Labeling (including the container label, package insert, and, if applicable, Medication Guide) proposed for the drug product must be the same as the

labeling approved for the reference listed drug, except for changes required because of differences approved under a petition filed under § 314.93 or because the drug product and the reference listed drug are produced or distributed by different manufacturers. Such differences between the applicant's proposed labeling and labeling approved for the reference listed drug may include differences in expiration date, formulation, bioavailability, or pharmacokinetics, labeling revisions made to comply with current FDA labeling guidelines or other guidance, or omission of an indication or other aspect of labeling protected by patent or accorded exclusivity under section 505(j)(4)(D) of the act.

(9) *Chemistry, manufacturing, and controls.* (i) The information required under § 314.50(d)(1), except that § 314.50(d)(1)(ii)(*c*) shall contain the proposed or actual master production record, including a description of the equipment, to be used for the manufacture of a commercial lot of the drug product.

(ii) *Inactive ingredients.* Unless otherwise stated in paragraphs (a)(9)(iii) through (a)(9)(v) of this section, an applicant shall identify and characterize the inactive ingredients in the proposed drug product and provide information demonstrating that such inactive ingredients do not affect the safety or efficacy of the proposed drug product.

(iii) *Inactive ingredient changes permitted in drug products intended for parenteral use.* Generally, a drug product intended for parenteral use shall contain the same inactive ingredients and in the same concentration as the reference listed drug identified by the applicant under paragraph (a)(3) of this section. However, an applicant may seek approval of a drug product that differs from the reference listed drug in preservative, buffer, or antioxidant provided that the applicant identifies and characterizes the differences and provides information demonstrating that the differences do not affect the safety or efficacy of the proposed drug product.

(iv) *Inactive ingredient changes permitted in drug products intended for ophthalmic or otic use.* Generally, a drug product intended for ophthalmic or otic use shall contain the same inactive ingredients and in the same concentration as the reference listed drug identified by the applicant under paragraph (a)(3) of this section. However, an applicant may seek approval of a drug product that differs from the reference listed drug in preservative, buffer, substance to adjust tonicity, or thickening agent provided that the applicant identifies and characterizes the differences and provides information demonstrating that the differences do not affect the safety or efficacy of the proposed drug product, except that, in a product intended for ophthalmic use, an applicant may not change a buffer or substance to adjust tonicity for the purpose of claiming a therapeutic advantage over or difference from the listed drug, e.g., by using a balanced salt solution as a diluent as opposed to an isotonic saline solution, or by making a significant change in the pH or other change that may raise questions of irritability.

(v) *Inactive ingredient changes permitted in drug products intended for topical use.* Generally, a drug product intended for topical use, solutions for aerosolization or nebulization, and nasal solutions shall contain the same inactive ingredients as the reference listed drug identified by the applicant under paragraph (a)(3) of this section. However, an abbreviated application may include different inactive ingredients provided that the applicant identifies and characterizes the differences and provides information demonstrating that the differences do not affect the safety or efficacy of the proposed drug product.

(10) *Samples.* The information required under § 314.50(e)(1) and (e)(2)(i). Samples need not be submitted until requested by FDA.

(11) *Other.* The information required under § 314.50(g).

(12) *Patent certification*—(i) *Patents claiming drug, drug product, or method of use.* (A) Except as provided in paragraph (a)(12)(iv) of this section, a certification with respect to each patent issued by the United States Patent and Trademark Office that, in the opinion of the applicant and to the best of its knowledge, claims the reference listed

drug or that claims a use of such listed drug for which the applicant is seeking approval under section 505(j) of the act and for which information is required to be filed under section 505(b) and (c) of the act and §314.53. For each such patent, the applicant shall provide the patent number and certify, in its opinion and to the best of its knowledge, one of the following circumstances:

(*1*) That the patent information has not been submitted to FDA. The applicant shall entitle such a certification "Paragraph I Certification";

(*2*) That the patent has expired. The applicant shall entitle such a certification "Paragraph II Certification";

(*3*) The date on which the patent will expire. The applicant shall entitle such a certification "Paragraph III Certification"; or

(*4*) That the patent is invalid, unenforceable, or will not be infringed by the manufacture, use, or sale of the drug product for which the abbreviated application is submitted. The applicant shall entitle such a certification "Paragraph IV Certification". This certification shall be submitted in the following form:

I, (*name of applicant*), certify that Patent No. _____ (*is invalid, unenforceable, or will not be infringed by the manufacture, use, or sale of*) (*name of proposed drug product*) for which this application is submitted.

The certification shall be accompanied by a statement that the applicant will comply with the requirements under §314.95(a) with respect to providing a notice to each owner of the patent or their representatives and to the holder of the approved application for the listed drug, and with the requirements under §314.95(c) with respect to the content of the notice.

(B) If the abbreviated new drug application refers to a listed drug that is itself a licensed generic product of a patented drug first approved under section 505(b) of the act, the appropriate patent certification under paragraph (a)(12)(i) of this section with respect to each patent that claims the first-approved patented drug or that claims a use for such drug.

(ii) *No relevant patents.* If, in the opinion of the applicant and to the best of its knowledge, there are no patents described in paragraph (a)(12)(i) of this section, a certification in the following form:

In the opinion and to the best knowledge of (*name of applicant*), there are no patents that claim the listed drug referred to in this application or that claim a use of the listed drug.

(iii) *Method of use patent.* (A) If patent information is submitted under section 505(b) or (c) of the act and §314.53 for a patent claiming a method of using the listed drug, and the labeling for the drug product for which the applicant is seeking approval does not include any indications that are covered by the use patent, a statement explaining that the method of use patent does not claim any of the proposed indications.

(B) If the labeling of the drug product for which the applicant is seeking approval includes an indication that, according to the patent information submitted under section 505(b) or (c) of the act and §314.53 or in the opinion of the applicant, is claimed by a use patent, an applicable certification under paragraph (a)(12)(i) of this section.

(iv) *Method of manufacturing patent.* An applicant is not required to make a certification with respect to any patent that claims only a method of manufacturing the listed drug.

(v) *Licensing agreements.* If the abbreviated new drug application is for a drug or method of using a drug claimed by a patent and the applicant has a licensing agreement with the patent owner, a certification under paragraph (a)(12)(i)(A)(*4*) of this section ("Paragraph IV Certification") as to that patent and a statement that it has been granted a patent license.

(vi) *Late submission of patent information.* If a patent on the listed drug is issued and the holder of the approved application for the listed drug does not submit the required information on the patent within 30 days of issuance of the patent, an applicant who submitted an abbreviated new drug application for that drug that contained an appropriate patent certification before the submission of the patent information is

not required to submit an amended certification. An applicant whose abbreviated new drug application is submitted after a late submission of patent information, or whose pending abbreviated application was previously submitted but did not contain an appropriate patent certification at the time of the patent submission, shall submit a certification under paragraph (a)(12)(i) of this section or a statement under paragraph (a)(12)(iii) of this section as to that patent.

(vii) *Disputed patent information.* If an applicant disputes the accuracy or relevance of patent information submitted to FDA, the applicant may seek a confirmation of the correctness of the patent information in accordance with the procedures under §314.53(f). Unless the patent information is withdrawn or changed, the applicant shall submit an appropriate certification for each relevant patent.

(viii) *Amended certifications.* A certification submitted under paragraphs (a)(12)(i) through (a)(12)(iii) of this section may be amended at any time before the effective date of the approval of the application. However, an applicant who has submitted a paragraph IV patent certification may not change it to a paragraph III certification if a patent infringement suit has been filed against another paragraph IV applicant unless the agency has determined that no applicant is entitled to 180-day exclusivity or the patent expires before the lawsuit is resolved or expires after the suit is resolved but before the end of the 180-day exclusivity period. If an applicant with a pending application voluntarily makes a patent certification for an untimely filed patent, the applicant may withdraw the patent certification for the untimely filed patent. An applicant shall submit an amended certification by letter or as an amendment to a pending application or by letter to an approved application. Once an amendment or letter is submitted, the application will no longer be considered to contain the prior certification.

(A) *After finding of infringement.* An applicant who has submitted a certification under paragraph (a)(12)(i)(A)(*4*) of this section and is sued for patent infringement within 45 days of the receipt of notice sent under §314.95 shall amend the certification if a final judgment in the action against the applicant is entered finding the patent to be infringed. In the amended certification, the applicant shall certify under paragraph (a)(12)(i)(A)(*3*) of this section that the patent will expire on a specific date. Once an amendment or letter for the change has been submitted, the application will no longer be considered to be one containing a certification under paragraph (a)(12)(i)(A)(*4*) of this section. If a final judgment finds the patent to be invalid and infringed, an amended certification is not required.

(B) *After removal of a patent from the list.* If a patent is removed from the list, any applicant with a pending application (including a tentatively approved application with a delayed effective date) who has made a certification with respect to such patent shall amend its certification. The applicant shall certify under paragraph (a)(12)(ii) of this section that no patents described in paragraph (a)(12)(i) of this section claim the drug or, if other relevant patents claim the drug, shall amend the certification to refer only to those relevant patents. In the amendment, the applicant shall state the reason for the change in certification (that the patent is or has been removed from the list). A patent that is the subject of a lawsuit under §314.107(c) shall not be removed from the list until FDA determines either that no delay in effective dates of approval is required under that section as a result of the lawsuit, that the patent has expired, or that any such period of delay in effective dates of approval is ended. An applicant shall submit an amended certification. Once an amendment or letter for the change has been submitted, the application will no longer be considered to be one containing a certification under paragraph (a)(12)(i)(A)(4) of this section.

(C) *Other amendments.* (*1*) Except as provided in paragraphs (a)(12)(vi) and (a)(12)(viii)(C)(*2*) of this section, an applicant shall amend a submitted certification if, at any time before the effective date of the approval of the application, the applicant learns that the submitted certification is no longer accurate.

(*2*) An applicant is not required to amend a submitted certification when information on a patent on the listed drug is submitted after the effective date of approval of the abbreviated application.

(13) *Financial certification or disclosure statement.* An abbreviated application shall contain a financial certification or disclosure statement as required by part 54 of this chapter.

(b) *Drug products subject to the Drug Efficacy Study Implementation (DESI) review.* If the abbreviated new drug application is for a duplicate of a drug product that is subject to FDA's DESI review (a review of drug products approved as safe between 1938 and 1962) or other DESI-like review and the drug product evaluated in the review is a listed drug, the applicant shall comply with the provisions of paragraph (a) of this section.

(c) [Reserved]

(d) *Format of an abbreviated application.* (1) The applicant shall submit a complete archival copy of the abbreviated application as required under paragraphs (a) and (c) of this section. FDA will maintain the archival copy during the review of the application to permit individual reviewers to refer to information that is not contained in their particular technical sections of the application, to give other agency personnel access to the application for official business, and to maintain in one place a complete copy of the application. An applicant may submit all or portions of the archival copy of the abbreviated application in any form (e.g., microfiche, optical disc, and magnetic tape) that the applicant and FDA agree is acceptable.

(2) For abbreviated new drug applications, the applicant shall submit a review copy of the abbreviated application that contains two separate sections. One section shall contain the information described under paragraphs (a)(2) through (a)(6), (a)(8), and (a)(9) of this section 505(j)(2)(A)(vii) of the act and one copy of the analytical methods and descriptive information needed by FDA's laboratories to perform tests on samples of the proposed drug product and to validate the applicant's analytical methods. The other section shall contain the information described under paragraphs (a)(3), (a)(7), and (a)(8) of this section. Each of the sections in the review copy is required to contain a copy of the application form described under §314.50(a).

(3) [Reserved]

(4) The applicant may obtain from FDA sufficient folders to bind the archival, the review, and the field copies of the abbreviated application.

(5) The applicant shall submit a field copy of the abbreviated application that contains the technical section described in paragraph (a)(9) of this section, a copy of the application form required under paragraph (a)(1) of this section, and a certification that the field copy is a true copy of the technical section described in paragraph (a)(9) of this section contained in the archival and review copies of the abbreviated application.

[57 FR 17983, Apr. 28, 1992; 57 FR 29353, July 1, 1992, as amended at 58 FR 47352, Sept. 8, 1993; 59 FR 50364, Oct. 3, 1994; 63 FR 5252, Feb. 2, 1998; 63 FR 66399, Dec. 1, 1998; 64 FR 401, Jan. 5, 1999; 65 FR 56479, Sept. 19, 2000; 67 FR 77672, Dec. 19, 2002]

EFFECTIVE DATE NOTE: At 68 FR 69019, Dec. 11, 2003, §314.94 was amended by revising paragraph (d)(1), effective June 8, 2004. For the convenience of the user, the revised text is set forth as follows:

§314.94 Content and format of an abbreviated application.

* * * * *

(d) * * * (1) The applicant must submit a complete archival copy of the abbreviated application as required under paragraphs (a) and (c) of this section. FDA will maintain the archival copy during the review of the application to permit individual reviewers to refer to information that is not contained in their particular technical sections of the application, to give other agency personnel access to the application for official business, and to maintain in one place a complete copy of the application.

(i) *Format of submission.* An applicant may submit portions of the archival copy of the abbreviated application in any form that the applicant and FDA agree is acceptable, except as provided in paragraph (d)(1)(ii) of this section.

(ii) *Labeling.* The content of labeling required under §201.100(d)(3) of this chapter (commonly referred to as the package insert or professional labeling), including all text, tables, and figures, must be submitted to the agency in electronic format as described in

paragraph (d)(1)(iii) of this section. This requirement applies to the content of labeling for the proposed drug product only and is in addition to the requirements of paragraph (a)(8)(ii) of this section that copies of the formatted label and all proposed labeling be submitted. Submissions under this paragraph must be made in accordance with part 11 of this chapter, except for the requirements of §11.10(a), (c) through (h), and (k), and the corresponding requirements of §11.30.

(iii) *Electronic format submissions.* Electronic format submissions must be in a form that FDA can process, review, and archive. FDA will periodically issue guidance on how to provide the electronic submission (e.g., method of transmission, media, file formats, preparation and organization of files).

* * * * *

§ 314.95 Notice of certification of invalidity or noninfringement of a patent.

(a) *Notice of certification.* For each patent that claims the listed drug or that claims a use for such listed drug for which the applicant is seeking approval and that the applicant certifies under §314.94(a)(12) is invalid, unenforceable, or will not be infringed, the applicant shall send notice of such certification by registered or certified mail, return receipt requested to each of the following persons:

(1) Each owner of the patent which is the subject of the certification or the representative designated by the owner to receive the notice. The name and address of the patent owner or its representative may be obtained from the United States Patent and Trademark Office; and

(2) The holder of the approved application under section 505(b) of the act for the listed drug that is claimed by the patent and for which the applicant is seeking approval, or, if the application holder does not reside or maintain a place of business within the United States, the application holder's attorney, agent, or other authorized official. The name and address of the application holder or its attorney, agent, or authorized official may be obtained from the Division of Drug Information Resources (HFD–80), Center for Drug Evaluation and Research, Food and Drug Administration, 5600 Fishers Lane, Rockville, MD 20857.

(3) This paragraph does not apply to a use patent that claims no uses for which the applicant is seeking approval.

(b) *Sending the notice.* The applicant shall send the notice required by paragraph (a) of this section when it receives from FDA an acknowledgment letter stating that its abbreviated new drug application is sufficiently complete to permit a substantive review. At the same time, the applicant shall amend its abbreviated new drug application to include a statement certifying that the notice has been provided to each person identified under paragraph (a) of this section and that the notice met the content requirements under paragraph (c) of this section.

(c) *Contents of a notice.* In the notice, the applicant shall cite section 505(j)(2)(B)(ii) of the act and shall include, but not be limited to, the following information:

(1) A statement that FDA has received an abbreviated new drug application submitted by the applicant containing any required bioavailability or bioequivalence data or information.

(2) The abbreviated application number.

(3) The established name, if any, as defined in section 502(e)(3) of the act, of the proposed drug product.

(4) The active ingredient, strength, and dosage form of the proposed drug product.

(5) The patent number and expiration date, as submitted to the agency or as known to the applicant, of each patent alleged to be invalid, unenforceable, or not infringed.

(6) A detailed statement of the factual and legal basis of the applicant's opinion that the patent is not valid, unenforceable, or will not be infringed. The applicant shall include in the detailed statement:

(i) For each claim of a patent alleged not to be infringed, a full and detailed explanation of why the claim is not infringed.

(ii) For each claim of a patent alleged to be invalid or unenforceable, a full and detailed explanation of the grounds supporting the allegation.

(7) If the applicant does not reside or have a place of business in the United States, the name and address of an

TITLE 21--FOOD AND DRUGS

CHAPTER 9--FEDERAL FOOD, DRUG, AND COSMETIC ACT

SUBCHAPTER V--DRUGS AND DEVICES

Part A--Drugs and Devices

Sec. 355a. Pediatric studies of drugs

(a) Definitions

As used in this section, the term ``pediatric studies'' or ``studies'' means at least one clinical investigation (that, at the Secretary's discretion, may include pharmacokinetic studies) in pediatric age groups (including neonates in appropriate cases) in which a drug is anticipated to be used.

(b) Market exclusivity for new drugs

If, prior to approval of an application that is submitted under section 355(b)(1) of this title, the Secretary determines that information relating to the use of a new drug in the pediatric population may produce health benefits in that population, the Secretary makes a written request for pediatric studies (which shall include a timeframe for completing such studies), and such studies are completed within any such timeframe and the reports thereof submitted in accordance with subsection (d)(2) of this section or accepted in accordance with subsection (d)(3) of this section--

(1)(A)(i) the period referred to in subsection (c)(3)(D)(ii) of section 355 of this title, and in subsection (j)(5)(D)(ii) of such section, is deemed to be five years and six months rather than five years, and the references in subsections (c)(3)(D)(ii) and (j)(5)(D)(ii) of such section to four years, to forty-eight months, and to seven and one-half years are deemed to be four and one-half years, fifty-four months, and eight years, respectively; or

(ii) the period referred to in clauses (iii) and (iv) of subsection (c)(3)(D) of such section, and in clauses (iii) and (iv) of subsection (j)(5)(D) of such section, is deemed to be three years and six months rather than three years; and

(B) if the drug is designated under section 360bb of this title for a rare disease or condition, the period referred to in section 360cc(a) of this title is deemed to be seven years and six months rather than seven years; and

(2)(A) if the drug is the subject of--

(i) a listed patent for which a certification has been submitted under subsection (b)(2)(A)(ii) or (j)(2)(A)(vii)(II) of section 355 of this title and for which pediatric studies were submitted prior to the expiration of the patent (including any patent extensions); or

(ii) a listed patent for which a certification has been

submitted under subsections (b)(2)(A)(iii) or (j)(2)(A)(vii)(III) of section 355 of this title,

the period during which an application may not be approved under section 355(c)(3) of this title or section 355(j)(4)(B) of this title shall be extended by a period of six months after the date the patent expires (including any patent extensions); or

(B) if the drug is the subject of a listed patent for which a certification has been submitted under subsection (b)(2)(A)(iv) or (j)(2)(A)(vii)(IV) of section 355 of this title, and in the patent infringement litigation resulting from the certification the court determines that the patent is valid and would be infringed, the period during which an application may not be approved under section 355(c)(3) of this title or section 355(j)(4)(B) of this title shall be extended by a period of six months after the date the patent expires (including any patent extensions).

(c) Market exclusivity for already-marketed drugs

If the Secretary determines that information relating to the use of an approved drug in the pediatric population may produce health benefits in that population and makes a written request to the holder of an approved application under section 355(b)(1) of this title for pediatric studies (which shall include a timeframe for completing such studies), the holder agrees to the request, the studies are completed within any such timeframe, and the reports thereof are submitted in accordance with subsection (d)(2) of this section or accepted in accordance with subsection (d)(3) of this section--

(1)(A)(i) the period referred to in subsection (c)(3)(D)(ii) of section 355 of this title, and in subsection (j)(5)(D)(ii) of such section, is deemed to be five years and six months rather than five years, and the references in subsections (c)(3)(D)(ii) and (j)(5)(D)(ii) of such section to four years, to forty-eight months, and to seven and one-half years are deemed to be four and one-half years, fifty-four months, and eight years, respectively; or

(ii) the period referred to in clauses (iii) and (iv) of subsection (c)(3)(D) of such section, and in clauses (iii) and (iv) of subsection (j)(5)(D) of such section, is deemed to be three years and six months rather than three years; and

(B) if the drug is designated under section 360bb of this title for a rare disease or condition, the period referred to in section 360cc(a) of this title is deemed to be seven years and six months rather than seven years; and

(2)(A) if the drug is the subject of--

(i) a listed patent for which a certification has been submitted under subsection (b)(2)(A)(ii) or (j)(2)(A)(vii)(II) of section 355 of this title and for which pediatric studies were submitted prior to the expiration of the patent (including any patent extensions); or

(ii) a listed patent for which a certification has been submitted under subsection (b)(2)(A)(iii) or (j)(2)(A)(vii)(III) of section 355 of this title,

the period during which an application may not be approved under section 355(c)(3) of this title or section 355(j)(4)(B) of this title shall be extended by a period of six months after the date the patent expires (including any patent extensions); or

(B) if the drug is the subject of a listed patent for which a certification has been submitted under subsection (b)(2)(A)(iv) or (j)(2)(A)(vii)(IV) of section 355 of this title, and in the patent

infringement litigation resulting from the certification the court determines that the patent is valid and would be infringed, the period during which an application may not be approved under section 355(c)(3) of this title or section 355(j)(4)(B) of this title shall be extended by a period of six months after the date the patent expires (including any patent extensions).

(d) Conduct of pediatric studies

(1) Agreement for studies

The Secretary may, pursuant to a written request from the Secretary under subsection (b) or (c) of this section, after consultation with--

(A) the sponsor of an application for an investigational new drug under section 355(i) of this title;

(B) the sponsor of an application for a new drug under section 355(b)(1) of this title; or

(C) the holder of an approved application for a drug under section 355(b)(1) of this title,

agree with the sponsor or holder for the conduct of pediatric studies for such drug. Such agreement shall be in writing and shall include a timeframe for such studies.

(2) Written protocols to meet the studies requirement

If the sponsor or holder and the Secretary agree upon written protocols for the studies, the studies requirement of subsection (b) or (c) of this section is satisfied upon the completion of the studies and submission of the reports thereof in accordance with the original written request and the written agreement referred to in paragraph (1). In reaching an agreement regarding written protocols, the Secretary shall take into account adequate representation of children of ethnic and racial minorities. Not later than 60 days after the submission of the report of the studies, the Secretary shall determine if such studies were or were not conducted in accordance with the original written request and the written agreement and reported in accordance with the requirements of the Secretary for filing and so notify the sponsor or holder.

(3) Other methods to meet the studies requirement

If the sponsor or holder and the Secretary have not agreed in writing on the protocols for the studies, the studies requirement of subsection (b) or (c) of this section is satisfied when such studies have been completed and the reports accepted by the Secretary. Not later than 90 days after the submission of the reports of the studies, the Secretary shall accept or reject such reports and so notify the sponsor or holder. The Secretary's only responsibility in accepting or rejecting the reports shall be to determine, within the 90 days, whether the studies fairly respond to the written request, have been conducted in accordance with commonly accepted scientific principles and protocols, and have been reported in accordance with the requirements of the Secretary for filing.

(4) Written request to holders of approved applications for drugs that have market exclusivity

(A) Request and response

If the Secretary makes a written request for pediatric studies (including neonates, as appropriate) under subsection (c) of this section to the holder of an application approved under section 355(b)(1) of this title, the holder, not later than 180 days after receiving the written request, shall respond to the Secretary as to the intention of the holder to act on the request by--

(i) indicating when the pediatric studies will be initiated, if the holder agrees to the request; or

(ii) indicating that the holder does not agree to the request.

(B) No agreement to request

(i) Referral

If the holder does not agree to a written request within the time period specified in subparagraph (A), and if the Secretary determines that there is a continuing need for information relating to the use of the drug in the pediatric population (including neonates, as appropriate), the Secretary shall refer the drug to the Foundation for the National Institutes of Health established under section 290b of title 42 (referred to in this paragraph as the ``Foundation'') for the conduct of the pediatric studies described in the written request.

(ii) Public notice

The Secretary shall give public notice of the name of the drug, the name of the manufacturer, and the indications to be studied made in a referral under clause (i).

(C) Lack of funds

On referral of a drug under subparagraph (B)(i), the Foundation shall issue a proposal to award a grant to conduct the requested studies unless the Foundation certifies to the Secretary, within a timeframe that the Secretary determines is appropriate through guidance, that the Foundation does not have funds available under section 290b(j)(9)(B)(i) \1\ of title 42 to conduct the requested studies. If the Foundation so certifies, the Secretary shall refer the drug for inclusion on the list established under section 284m of title 42 for the conduct of the studies.

\1\ See References in Text note below.

(D) Effect of subsection

Nothing in this subsection (including with respect to referrals from the Secretary to the Foundation) alters or amends section 331(j) of this title or section 552 of title 5 or section 1905 of title 18.

(E) No requirement to refer

Nothing in this subsection shall be construed to require that every declined written request shall be referred to the

Foundation.

(F) Written requests under subsection (b)

For drugs under subsection (b) of this section for which written requests have not been accepted, if the Secretary determines that there is a continuing need for information relating to the use of the drug in the pediatric population (including neonates, as appropriate), the Secretary shall issue a written request under subsection (c) of this section after the date of approval of the drug.

(e) Delay of effective date for certain application

If the Secretary determines that the acceptance or approval of an application under section 355(b)(2) or 355(j) of this title for a new drug may occur after submission of reports of pediatric studies under this section, which were submitted prior to the expiration of the patent (including any patent extension) or the applicable period under clauses (ii) through (iv) of section 355(c)(3)(D) of this title or clauses (ii) through (iv) of section 355(j)(5)(D) of this title, but before the Secretary has determined whether the requirements of subsection (d) of this section have been satisfied, the Secretary shall delay the acceptance or approval under section 355(b)(2) or 355(j) of this title until the determination under subsection (d) of this section is made, but any such delay shall not exceed 90 days. In the event that requirements of this section are satisfied, the applicable six-month period under subsection (b) or (c) of this section shall be deemed to have been running during the period of delay.

(f) Notice of determinations on studies requirement

The Secretary shall publish a notice of any determination that the requirements of subsection (d) of this section have been met and that submissions and approvals under subsection (b)(2) or (j) of section 355 of this title for a drug will be subject to the provisions of this section.

(g) Limitations

A drug to which the six-month period under subsection (b) or (c) of this section has already been applied--

(1) may receive an additional six-month period under subsection (c)(1)(A)(ii) of this section for a supplemental application if all other requirements under this section are satisfied, except that such a drug may not receive any additional such period under subsection (c)(2) of this section; and

(2) may not receive any additional such period under subsection (c)(1)(B) of this section.

(h) Relationship to regulations

Notwithstanding any other provision of law, if any pediatric study is required pursuant to regulations promulgated by the Secretary and such study meets the completeness, timeliness, and other requirements of this section, such study shall be deemed to satisfy the requirement for market exclusivity pursuant to this section.

(i) Labeling supplements

(1) Priority status for pediatric supplements

Any supplement to an application under section 355 of this title proposing a labeling change pursuant to a report on a pediatric study under this section--

(A) shall be considered to be a priority supplement; and

(B) shall be subject to the performance goals established by the Commissioner for priority drugs.

(2) Dispute resolution

(A) Request for labeling change and failure to agree

If the Commissioner determines that an application with respect to which a pediatric study is conducted under this section is approvable and that the only open issue for final action on the application is the reaching of an agreement between the sponsor of the application and the Commissioner on appropriate changes to the labeling for the drug that is the subject of the application, not later than 180 days after the date of submission of the application--

(i) the Commissioner shall request that the sponsor of the application make any labeling change that the Commissioner determines to be appropriate; and

(ii) if the sponsor of the application does not agree to make a labeling change requested by the Commissioner, the Commissioner shall refer the matter to the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee.

(B) Action by the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee

Not later than 90 days after receiving a referral under subparagraph (A)(ii), the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee shall--

(i) review the pediatric study reports; and

(ii) make a recommendation to the Commissioner concerning appropriate labeling changes, if any.

(C) Consideration of recommendations

The Commissioner shall consider the recommendations of the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee and, if appropriate, not later than 30 days after receiving the recommendation, make a request to the sponsor of the application to make any labeling change that the Commissioner determines to be appropriate.

(D) Misbranding

If the sponsor of the application, within 30 days after receiving a request under subparagraph (C), does not agree to make a labeling change requested by the Commissioner, the Commissioner may deem the drug that is the subject of the application to be misbranded.

(E) No effect on authority

Nothing in this subsection limits the authority of the

United States to bring an enforcement action under this chapter when a drug lacks appropriate pediatric labeling. Neither course of action (the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee process or an enforcement action referred to in the preceding sentence) shall preclude, delay, or serve as the basis to stay the other course of action.

(j) Dissemination of pediatric information

(1) In general

Not later than 180 days after the date of submission of a report on a pediatric study under this section, the Commissioner shall make available to the public a summary of the medical and clinical pharmacology reviews of pediatric studies conducted for the supplement, including by publication in the Federal Register.

(2) Effect of subsection

Nothing in this subsection alters or amends section 331(j) of this title or section 552 of title 5 or section 1905 of title 18.

(k) Clarification of interaction of market exclusivity under this section and market exclusivity awarded to an applicant for approval of a drug under section 355(j) of this title

If a 180-day period under section 355(j)(5)(B)(iv) of this title overlaps with a 6-month exclusivity period under this section, so that the applicant for approval of a drug under section 355(j) of this title entitled to the 180-day period under that section loses a portion of the 180-day period to which the applicant is entitled for the drug, the 180-day period shall be extended from--

(1) the date on which the 180-day period would have expired by the number of days of the overlap, if the 180-day period would, but for the application of this subsection, expire after the 6-month exclusivity period; or

(2) the date on which the 6-month exclusivity period expires, by the number of days of the overlap if the 180-day period would, but for the application of this subsection, expire during the six-month exclusivity period.

(l) Prompt approval of drugs under section 355(j) of this title when pediatric information is added to labeling

(1) General rule

A drug for which an application has been submitted or approved under section 355(j) of this title shall not be considered ineligible for approval under that section or misbranded under section 352 of this title on the basis that the labeling of the drug omits a pediatric indication or any other aspect of labeling pertaining to pediatric use when the omitted indication or other aspect is protected by patent or by exclusivity under clause (iii) or (iv) of section 355(j)(5)(D) of this title.

(2) Labeling

Notwithstanding clauses (iii) and (iv) of section 355(j)(5)(D) of this title, the Secretary may require that the labeling of a drug approved under section 355(j) of this title that omits a pediatric

indication or other aspect of labeling as described in paragraph (1) include--

(A) a statement that, because of marketing exclusivity for a manufacturer--

(i) the drug is not labeled for pediatric use; or

(ii) in the case of a drug for which there is an additional pediatric use not referred to in paragraph (1), the drug is not labeled for the pediatric use under paragraph (1); and

(B) a statement of any appropriate pediatric contraindications, warnings, or precautions that the Secretary considers necessary.

(3) Preservation of pediatric exclusivity and other provisions

This subsection does not affect--

(A) the availability or scope of exclusivity under this section;

(B) the availability or scope of exclusivity under section 355 of this title for pediatric formulations;

(C) the question of the eligibility for approval of any application under section 355(j) of this title that omits any other conditions of approval entitled to exclusivity under clause (iii) or (iv) of section 355(j)(5)(D) of this title; or

(D) except as expressly provided in paragraphs (1) and (2), the operation of section 355 of this title.

(m) Report

The Secretary shall conduct a study and report to Congress not later than January 1, 2001, based on the experience under the program established under this section. The study and report shall examine all relevant issues, including--

(1) the effectiveness of the program in improving information about important pediatric uses for approved drugs;

(2) the adequacy of the incentive provided under this section;

(3) the economic impact of the program on taxpayers and consumers, including the impact of the lack of lower cost generic drugs on patients, including on lower income patients; and

(4) any suggestions for modification that the Secretary determines to be appropriate.

(n) Sunset

A drug may not receive any 6-month period under subsection (b) or (c) of this section unless--

(1) on or before October 1, 2007, the Secretary makes a written request for pediatric studies of the drug;

(2) on or before October 1, 2007, an application for the drug is accepted for filing under section 355(b) of this title; and

(3) all requirements of this section are met.

(June 25, 1938, ch. 675, Sec. 505A, as added Pub. L. 105-115, title I, Sec. 111, Nov. 21, 1997, 111 Stat. 2305; amended Pub. L. 107-109, Secs. 2, 4, 5(b)(2), 7-11(a), 18(a), 19, Jan. 4, 2002, 115 Stat. 1408, 1411, 1413-1415, 1423, 1424.)

References in Text

Section 290b(j)(9)(B)(i) of title 42, referred to in subsec. (d)(4)(C), was in the original ``section 499(j)(9)(B)(i)'' and was translated as meaning section 499(j)(9)(B)(i) of the Public Health Service Act to reflect the probable intent of Congress because there is no section 499 of the Federal Food, Drug, and Cosmetic Act and section 499 of the Public Health Service Act relates to the establishment and duties of the National Foundation for Biomedical Research.

Amendments

2002--Subsec. (a). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (g) as (a). Former subsec. (a) redesignated (b).

Subsec. (a)(1)(A). Pub. L. 107-109, Sec. 19(1)(A), (B), substituted ``(j)(5)(D)(ii)'' for ``(j)(4)(D)(ii)'' in two places in cl. (i) and ``(j)(5)(D)'' for ``(j)(4)(D)'' in cl. (ii).

Subsec. (b). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (a) as (b).

Pub. L. 107-109, Sec. 2(1), struck out heading and text of subsec. (b). Text read as follows: ``Not later than 180 days after November 21, 1997, the Secretary, after consultation with experts in pediatric research shall develop, prioritize, and publish an initial list of approved drugs for which additional pediatric information may produce health benefits in the pediatric population. The Secretary shall annually update the list.''

Subsec. (c). Pub. L. 107-109, Sec. 2(2), in introductory provisions, inserted ``determines that information relating to the use of an approved drug in the pediatric population may produce health benefits in that population and'' after ``the Secretary'' and struck out ``concerning a drug identified in the list described in subsection (b) of this section'' after ``such studies)''.

Subsec. (c)(1)(A). Pub. L. 107-109, Sec. 19(1)(A), (B), substituted ``(j)(5)(D)(ii)'' for ``(j)(4)(D)(ii)'' in two places in cl. (i) and ``(j)(5)(D)'' for ``(j)(4)(D)'' in cl. (ii).

Subsec. (d)(1). Pub. L. 107-109, Sec. 19(4), substituted ``subsection (b) or (c)'' for ``subsection (a) or (c)'' in introductory provisions.

Subsec. (d)(2). Pub. L. 107-109, Secs. 18(a), 19(4), substituted ``subsection (b) or (c)'' for ``subsection (a) or (c)'' and inserted ``In reaching an agreement regarding written protocols, the Secretary shall take into account adequate representation of children of ethnic and racial minorities.'' after first sentence.

Subsec. (d)(3). Pub. L. 107-109, Sec. 19(4), substituted ``subsection (b) or (c)'' for ``subsection (a) or (c)''.

Subsec. (d)(4). Pub. L. 107-109, Sec. 4, added par. (4).

Subsec. (e). Pub. L. 107-109, Sec. 19(1)(C), (4), substituted ``section 355(j)(5)(D)'' for ``section 355(j)(4)(D)'' and ``subsection (b) or (c)'' for ``subsection (a) or (c)''.

Subsec. (g). Pub. L. 107-109, Sec. 19(2), (3), (5), redesignated subsec. (h) as (g) and substituted ``subsection (b) or (c)'' for ``subsection (a) or (b)'' in introductory provisions. Former subsec. (g) redesignated (a).

Pub. L. 107-109, Sec. 7, inserted ``(including neonates in appropriate cases)'' after ``pediatric age groups''.

Subsec. (h). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (i) as (h). Former subsec. (h) redesignated (g).

Subsec. (i). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (l) as (i). Former subsec. (i) redesignated (h).

Subsec. (j). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec.

(m) as (j). Former subsec. (j) redesignated (n).

Pub. L. 107-109, Sec. 8, added subsec. (j) and struck out heading and text of former subsec. (j). Text read as follows: ``A drug may not receive any six-month period under subsection (a) or (c) of this section unless the application for the drug under section 355(b)(1) of this title is submitted on or before January 1, 2002. After January 1, 2002, a drug shall receive a six-month period under subsection (c) of this section if--

``(1) the drug was in commercial distribution as of November 21, 1997;

``(2) the drug was included by the Secretary on the list under subsection (b) of this section as of January 1, 2002;

``(3) the Secretary determines that there is a continuing need for information relating to the use of the drug in the pediatric population and that the drug may provide health benefits in that population; and

``(4) all requirements of this section are met.''

Subsec. (k). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (n) as (k). Former subsec. (k) redesignated (m).

Subsec. (l). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (o) as (l). Former subsec. (l) redesignated (i).

Pub. L. 107-109, Sec. 5(b)(2), added subsec. (l).

Subsec. (m). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (k) as (m). Former subsec. (m) redesignated (j).

Pub. L. 107-109, Sec. 9, added subsec. (m).

Subsec. (n). Pub. L. 107-109, Sec. 19(4), which directed substitution of ``subsection (b) or (c)'' for ``subsection (a) or (c)'' in subsec. (m), was executed by making the substitution in introductory provisions of subsec. (n), to reflect the probable intent of Congress.

Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (j) as (n). Former subsec. (n) redesignated (k).

Pub. L. 107-109, Sec. 10, added subsec. (n).

Subsec. (o). Pub. L. 107-109, Sec. 19(2), (3), redesignated subsec. (o) as (l).

Pub. L. 107-109, Sec. 11(a), added subsec. (o).

Effective Date of 2002 Amendment

Pub. L. 107-109, Sec. 11(b), Jan. 4, 2002, 115 Stat. 1416, provided that: ``The amendment made by subsection (a) [amending this section] takes effect on the date of enactment of this Act [Jan. 4, 2002], including with respect to applications under section 505(j) of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355(j)) that are approved or pending on that date.''

Report on Pediatric Exclusivity Program

Pub. L. 107-109, Sec. 16, Jan. 4, 2002, 115 Stat. 1421, provided that: ``Not later than October 1, 2006, the Comptroller General of the United States, in consultation with the Secretary of Health and Human Services, shall submit to Congress a report that addresses the following issues, using publicly available data or data otherwise available to the Government that may be used and disclosed under applicable law:

``(1) The effectiveness of section 505A of the Federal Food, Drug, and Cosmetic Act [21 U.S.C. 355a] and section 409I of the Public Health Service Act [42 U.S.C. 284m] (as added by this Act) in ensuring that medicines used by children are tested and properly labeled, including--

``(A) the number and importance of drugs for children that are being tested as a result of this legislation and the importance for children, health care providers, parents, and others of labeling changes made as a result of such testing;

``(B) the number and importance of drugs for children that are not being tested for their use notwithstanding the provisions of this legislation, and possible reasons for the lack of testing; and

``(C) the number of drugs for which testing is being done, exclusivity granted, and labeling changes required, including the date pediatric exclusivity is granted and the date labeling changes are made and which labeling changes required the use of the dispute resolution process established pursuant to the amendments made by this Act [see Short Title of 2002 Amendment note set out under section 301 of this title], together with a description of the outcomes of such process, including a description of the disputes and the recommendations of the Pediatric Advisory Subcommittee of the Anti-Infective Drugs Advisory Committee.

``(2) The economic impact of section 505A of the Federal Food, Drug, and Cosmetic Act and section 409I of the Public Health Service Act (as added by this Act), including an estimate of--

``(A) the costs to taxpayers in the form of higher expenditures by medicaid and other Government programs;

``(B) sales for each drug during the 6-month period for which exclusivity is granted, as attributable to such exclusivity;

``(C) costs to consumers and private insurers as a result of any delay in the availability of lower cost generic equivalents of drugs tested and granted exclusivity under the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 301 et seq.), and loss of revenue by the generic drug industry and retail pharmacies as a result of any such delay; and

``(D) the benefits to the government, to private insurers, and to consumers resulting from decreased health care costs, including--

``(i) decreased hospitalizations and fewer medical errors, due to more appropriate and more effective use of medications in children as a result of testing and re-labeling because of the amendments made by this Act;

``(ii) direct and indirect benefits associated with fewer physician visits not related to hospitalization;

``(iii) benefits to children from missing less time at school and being less affected by chronic illnesses, thereby allowing a better quality of life;

``(iv) benefits to consumers from lower health insurance premiums due to lower treatment costs and hospitalization rates; and

``(v) benefits to employers from reduced need for employees to care for family members.

``(3) The nature and type of studies in children for each drug granted exclusivity under the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 301 et seq.), including--

``(A) a description of the complexity of the studies;

``(B) the number of study sites necessary to obtain appropriate data;

``(C) the number of children involved in any clinical studies; and

``(D) the estimated cost of each of the studies.

``(4) Any recommendations for modifications to the programs

established under section 505A of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355a) and section 409I of the Public Health Service Act [42 U.S.C. 284m] (as added by section 3) that the Secretary determines to be appropriate, including a detailed rationale for each recommendation.

``(5) The increased private and Government-funded pediatric research capability associated with this Act and the amendments made by this Act.

``(6) The number of written requests and additional letters of recommendation that the Secretary issues.

``(7) The prioritized list of off-patent drugs for which the Secretary issues written requests.

``(8)(A) The efforts made by the Secretary to increase the number of studies conducted in the neonate population; and

``(B) the results of those efforts, including efforts made to encourage the conduct of appropriate studies in neonates by companies with products that have sufficient safety and other information to make the conduct of studies ethical and safe.''

Study by General Accounting Office

Pub. L. 107-109, Sec. 18(b), Jan. 4, 2002, 115 Stat. 1423, provided that:

``(1) In general.--The Comptroller General of the United States shall conduct a study for the purpose of determining the following:

``(A) The extent to which children of ethnic and racial minorities are adequately represented in studies under section 505A of the Federal Food, Drug, and Cosmetic Act [21 U.S.C. 355a]; and to the extent ethnic and racial minorities are not adequately represented, the reasons for such under representation and recommendations to increase such representation.

``(B) Whether the Food and Drug Administration has appropriate management systems to monitor the representation of the children of ethnic and racial minorities in such studies.

``(C) Whether drugs used to address diseases that disproportionately affect racial and ethnic minorities are being studied for their safety and effectiveness under section 505A of the Federal Food, Drug, and Cosmetic Act.

``(2) Date certain for completing study.--Not later than January 10, 2003, the Comptroller General shall complete the study required in paragraph (1) and submit to the Congress a report describing the findings of the study.''

Section Referred to in Other Sections

This section is referred to in section 355b of this title; title 42 sections 284m, 290b.

PART 320—BIOAVAILABILITY AND BIOEQUIVALENCE REQUIREMENTS

Subpart A—General Provisions

Sec.
320.1 Definitions.

Subpart B—Procedures for Determining the Bioavailability or Bioequivalence of Drug Products

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320.63 Retention of bioequivalence samples.

AUTHORITY: 21 U.S.C. 321, 351, 352, 355, 371.

Subpart A—General Provisions

§ 320.1 Definitions.

(a) *Bioavailability* means the rate and extent to which the active ingredient or active moiety is absorbed from a drug product and becomes available at the site of action. For drug products that are not intended to be absorbed into the bloodstream, bioavailability may be assessed by measurements intended to reflect the rate and extent to which the active ingredient or active moiety becomes available at the site of action.

(b) *Drug product* means a finished dosage form, e.g., tablet, capsule, or solution, that contains the active drug ingredient, generally, but not necessarily, in association with inactive ingredients.

(c) *Pharmaceutical equivalents* means drug products in identical dosage forms that contain identical amounts of the identical active drug ingredient, i.e., the same salt or ester of the same therapeutic moiety, or, in the case of modified release dosage forms that require a reservoir or overage or such forms as prefilled syringes where residual volume may vary, that deliver identical amounts of the active drug ingredient over the identical dosing period; do not necessarily contain the same inactive ingredients; and meet the identical compendial or other applicable standard of identity, strength, quality, and purity, including potency and, where applicable, content uniformity, disintegration times, and/or dissolution rates.

(d) *Pharmaceutical alternatives* means drug products that contain the identical therapeutic moiety, or its precursor, but not necessarily in the same amount or dosage form or as the same salt or ester. Each such drug product individually meets either the identical or its own respective compendial or other applicable standard of identity, strength, quality, and purity, including potency and, where applicable, content uniformity, disintegration times and/or dissolution rates.

(e) *Bioequivalence* means the absence of a significant difference in the rate and extent to which the active ingredient or active moiety in pharmaceutical equivalents or pharmaceutical alternatives becomes available at the site of drug action when administered at the same molar dose under similar conditions in an appropriately designed study. Where there is an intentional difference in rate (e.g., in certain extended release dosage forms), certain

pharmaceutical equivalents or alternatives may be considered bioequivalent if there is no significant difference in the extent to which the active ingredient or moiety from each product becomes available at the site of drug action. This applies only if the difference in the rate at which the active ingredient or moiety becomes available at the site of drug action is intentional and is reflected in the proposed labeling, is not essential to the attainment of effective body drug concentrations on chronic use, and is considered medically insignificant for the drug.

(f) *Bioequivalence requirement* means a requirement imposed by the Food and Drug Administration for in vitro and/or in vivo testing of specified drug products which must be satisfied as a condition of marketing.

[42 FR 1634, Jan. 7, 1977, as amended at 42 FR 1648, Jan. 7, 1977; 57 FR 17997, Apr. 28, 1992; 67 FR 77672, Dec. 19, 2002]

Subpart B—Procedures for Determining the Bioavailability or Bioequivalence of Drug Products

SOURCE: 42 FR 1648, Jan. 7, 1977, unless otherwise noted.

§ 320.21 Requirements for submission of in vivo bioavailability and bioequivalence data.

(a) Any person submitting a full new drug application to the Food and Drug Administration (FDA) shall include in the application either:

(1) Evidence measuring the in vivo bioavailability of the drug product that is the subject of the application; or

(2) Information to permit FDA to waive the submission of evidence measuring in vivo bioavailability.

(b) Any person submitting an abbreviated new drug application to FDA shall include in the application either:

(1) Evidence demonstrating that the drug product that is the subject of the abbreviated new drug application is bioequivalent to the reference listed drug (defined in § 314.3(b) of this chapter); or

(2) Information to show that the drug product is bioequivalent to the reference listed drug which would permit FDA to waive the submission of evidence demonstrating in vivo bioequivalence as provided in paragraph (f) of this section.

(c) Any person submitting a supplemental application to FDA shall include in the supplemental application the evidence or information set forth in paragraphs (a) and (b) of this section if the supplemental application proposes any of the following changes:

(1) A change in the manufacturing site or a change in the manufacturing process, including a change in product formulation or dosage strength, beyond the variations provided for in the approved application.

(2) A change in the labeling to provide for a new indication for use of the drug product, if clinical studies are required to support the new indication for use.

(3) A change in the labeling to provide for a new dosage regimen or for an additional dosage regimen for a special patient population, e.g., infants, if clinical studies are required to support the new or additional dosage regimen.

(d) FDA may approve a full new drug application, or a supplemental application proposing any of the changes set forth in paragraph (c) of this section, that does not contain evidence of in vivo bioavailability or information to permit waiver of the requirement for in vivo bioavailability data, if all of the following conditions are met.

(1) The application is otherwise approvable.

(2) The application agrees to submit, within the time specified by FDA, either:

(i) Evidence measuring the in vivo bioavailability and demonstrating the in vivo bioequivalence of the drug product that is the subject of the application; or

(ii) Information to permit FDA to waive measurement of in vivo bioavailability.

(e) Evidence measuring the in vivo bioavailability and demonstrating the in vivo bioequivalence of a drug product shall be obtained using one of the approaches for determining bioavailability set forth in § 320.24.

(f) Information to permit FDA to waive the submission of evidence measuring the in vivo bioavailability or

differences in rate and extent of absorption that are not attributable to subject variability.

(3) A drug product that differs from the reference material in its rate of absorption, but not in its extent of absorption, may be considered to be bioavailable if the difference in the rate of absorption is intentional, is appropriately reflected in the labeling, is not essential to the attainment of effective body drug concentrations on chronic use, and is considered medically insignificant for the drug product.

(b) Two drug products will be considered bioequivalent drug products if they are pharmaceutical equivalents or pharmaceutical alternatives whose rate and extent of absorption do not show a significant difference when administered at the same molar dose of the active moiety under similar experimental conditions, either single dose or multiple dose. Some pharmaceutical equivalents or pharmaceutical alternatives may be equivalent in the extent of their absorption but not in their rate of absorption and yet may be considered bioequivalent because such differences in the rate of absorption are intentional and are reflected in the labeling, are not essential to the attainment of effective body drug concentrations on chronic use, and are considered medically insignificant for the particular drug product studied.

[57 FR 17999, Apr. 28, 1992, as amended at 67 FR 77673, Dec. 19, 2002]

§ 320.24 Types of evidence to measure bioavailability or establish bioequivalence.

(a) Bioavailability may be measured or bioequivalence may be demonstrated by several in vivo and in vitro methods. FDA may require in vivo or in vitro testing, or both, to measure the bioavailability of a drug product or establish the bioequivalence of specific drug products. Information on bioequivalence requirements for specific products is included in the current edition of FDA's publication "Approved Drug Products with Therapeutic Equivalence Evaluations" and any current supplement to the publication. The selection of the method used to meet an in vivo or in vitro testing requirement depends upon the purpose of the study, the analytical methods available, and the nature of the drug product. Applicants shall conduct bioavailability and bioequivalence testing using the most accurate, sensitive, and reproducible approach available among those set forth in paragraph (b) of this section. The method used must be capable of measuring bioavailability or establishing bioequivalence, as appropriate, for the product being tested.

(b) The following in vivo and in vitro approaches, in descending order of accuracy, sensitivity, and reproducibility, are acceptable for determining the bioavailability or bioequivalence of a drug product.

(1)(i) An in vivo test in humans in which the concentration of the active ingredient or active moiety, and, when appropriate, its active metabolite(s), in whole blood, plasma, serum, or other appropriate biological fluid is measured as a function of time. This approach is particularly applicable to dosage forms intended to deliver the active moiety to the bloodstream for systemic distribution within the body; or

(ii) An in vitro test that has been correlated with and is predictive of human in vivo bioavailability data; or

(2) An in vivo test in humans in which the urinary excretion of the active moiety, and, when appropriate, its active metabolite(s), are measured as a function of time. The intervals at which measurements are taken should ordinarily be as short as possible so that the measure of the rate of elimination is as accurate as possible. Depending on the nature of the drug product, this approach may be applicable to the category of dosage forms described in paragraph (b)(1)(i) of this section. This method is not appropriate where urinary excretion is not a significant mechanism of elimination.

(3) An in vivo test in humans in which an appropriate acute pharmacological effect of the active moiety, and, when appropriate, its active metabolite(s), are measured as a function of time if such effect can be measured with sufficient accuracy, sensitivity, and reproducibility. This approach is applicable to the category of dosage forms described in paragraph (b)(1)(i) of this section only when appropriate

methods are not available for measurement of the concentration of the moiety, and, when appropriate, its active metabolite(s), in biological fluids or excretory products but a method is available for the measurement of an appropriate acute pharmacological effect. This approach may be particularly applicable to dosage forms that are not intended to deliver the active moiety to the bloodstream for systemic distribution.

(4) Well-controlled clinical trials that establish the safety and effectiveness of the drug product, for purposes of measuring bioavailability, or appropriately designed comparative clinical trials, for purposes of demonstrating bioequivalence. This approach is the least accurate, sensitive, and reproducible of the general approaches for measuring bioavailability or demonstrating bioequivalence. For dosage forms intended to deliver the active moiety to the bloodstream for systemic distribution, this approach may be considered acceptable only when analytical methods cannot be developed to permit use of one of the approaches outlined in paragraphs (b)(1)(i) and (b)(2) of this section, when the approaches described in paragraphs (b)(1)(ii), (b)(1)(iii), and (b)(3) of this section are not available. This approach may also be considered sufficiently accurate for measuring bioavailability or demonstrating bioequivalence of dosage forms intended to deliver the active moiety locally, e.g., topical preparations for the skin, eye, and mucous membranes; oral dosage forms not intended to be absorbed, e.g., an antacid or radiopaque medium; and bronchodilators administered by inhalation if the onset and duration of pharmacological activity are defined.

(5) A currently available in vitro test acceptable to FDA (usually a dissolution rate test) that ensures human in vivo bioavailability.

(6) Any other approach deemed adequate by FDA to measure bioavailability or establish bioequivalence.

(c) FDA may, notwithstanding prior requirements for measuring bioavailability or establishing bioequivalence, require in vivo testing in humans of a product at any time if the agency has evidence that the product:

(1) May not produce therapeutic effects comparable to a pharmaceutical equivalent or alternative with which it is intended to be used interchangeably;

(2) May not be bioequivalent to a pharmaceutical equivalent or alternative with which it is intended to be used interchangeably; or

(3) Has greater than anticipated potential toxicity related to pharmacokinetic or other characteristics.

[57 FR 17999, Apr. 28, 1992; 57 FR 29354, July 1, 1992, as amended at 67 FR 77673, Dec. 19, 2002]

§ 320.25 Guidelines for the conduct of an in vivo bioavailability study.

(a) *Guiding principles.* (1) The basic principle in an in vivo bioavailability study is that no unnecessary human research should be done.

(2) An in vivo bioavailability study is generally done in a normal adult population under standardized conditions. In some situations, an in vivo bioavailability study in humans may preferably and more properly be done in suitable patients. Critically ill patients shall not be included in an in vivo bioavailability study unless the attending physician determines that there is a potential benefit to the patient.

(b) *Basic design.* The basic design of an in vivo bioavailability study is determined by the following:

(1) The scientific questions to be answered.

(2) The nature of the reference material and the dosage form to be tested.

(3) The availability of analytical methods.

(4) Benefit-risk considerations in regard to testing in humans.

(c) *Comparison to a reference material.* In vivo bioavailability testing of a drug product shall be in comparison to an appropriate reference material unless some other approach is more appropriate for valid scientific reasons.

(d) *Previously unmarketed active drug ingredients or therapeutic moieties.* (1) An in vivo bioavailability study involving a drug product containing an active drug ingredient or therapeutic moiety that has not been approved for marketing can be used to measure the following pharmacokinetic data:

demonstrating the in vivo bioequivalence shall meet the criteria set forth in § 320.22.

(g) Any person holding an approved full or abbreviated new drug application shall submit to FDA a supplemental application containing new evidence measuring the in vivo bioavailability or demonstrating the in vivo bioequivalence of the drug product that is the subject of the application if notified by FDA that:

(1) There are data demonstrating that the dosage regimen in the labeling is based on incorrect assumptions or facts regarding the pharmacokinetics of the drug product and that following this dosage regimen could potentially result in subtherapeutic or toxic levels; or

(2) There are data measuring significant intra-batch and batch-to-batch variability, e.g., plus or minus 25 percent, in the bioavailability of the drug product.

(h) The requirements of this section regarding the submission of evidence measuring the in vivo bioavailability or demonstrating the in vivo bioequivalence apply only to a full or abbreviated new drug application or a supplemental application for a finished dosage formulation.

[57 FR 17998, Apr. 28, 1992, as amended at 67 FR 77672, Dec. 19, 2002]

§ 320.22 Criteria for waiver of evidence of in vivo bioavailability or bioequivalence.

(a) Any person submitting a full or abbreviated new drug application, or a supplemental application proposing any of the changes set forth in § 320.21(c), may request FDA to waive the requirement for the submission of evidence measuring the in vivo bioavailability or demonstrating the in vivo bioequivalence of the drug product that is the subject of the application. An applicant shall submit a request for waiver with the application. Except as provided in paragraph (f) of this section, FDA shall waive the requirement for the submission of evidence of in vivo bioavailability or bioequivalence if the drug product meets any of the provisions of paragraphs (b), (c), (d), or (e) of this section.

(b) For certain drug products, the in vivo bioavailability or bioequivalence of the drug product may be self-evident. FDA shall waive the requirement for the submission of evidence obtained in vivo measuring the bioavailability or demonstrating the bioequivalence of these drug products. A drug product's in vivo bioavailability or bioequivalence may be considered self-evident based on other data in the application if the product meets one of the following criteria:

(1) The drug product:

(i) Is a parenteral solution intended solely for administration by injection, or an ophthalmic or otic solution; and

(ii) Contains the same active and inactive ingredients in the same concentration as a drug product that is the subject of an approved full new drug application or abbreviated new drug application.

(2) The drug product:

(i) Is administered by inhalation as a gas, e.g., a medicinal or an inhalation anesthetic; and

(ii) Contains an active ingredient in the same dosage form as a drug product that is the subject of an approved full new drug application or abbreviated new drug application.

(3) The drug product:

(i) Is a solution for application to the skin, an oral solution, elixir, syrup, tincture, a solution for aerosolization or nebulization, a nasal solution, or similar other solubilized form; and

(ii) Contains an active drug ingredient in the same concentration and dosage form as a drug product that is the subject of an approved full new drug application or abbreviated new drug application; and

(iii) Contains no inactive ingredient or other change in formulation from the drug product that is the subject of the approved full new drug application or abbreviated new drug application that may significantly affect absorption of the active drug ingredient or active moiety for products that are systemically absorbed, or that may significantly affect systemic or local availability for products intended to act locally.

(c) FDA shall waive the requirement for the submission of evidence measuring the in vivo bioavailability or

demonstrating the in vivo bioequivalence of a solid oral dosage form (other than a delayed release or extended release dosage form) of a drug product determined to be effective for at least one indication in a Drug Efficacy Study Implementation notice or which is identical, related, or similar to such a drug product under § 310.6 of this chapter unless FDA has evaluated the drug product under the criteria set forth in § 320.33, included the drug product in the Approved Drug Products with Therapeutic Equivalence Evaluations List, and rated the drug product as having a known or potential bioequivalence problem. A drug product so rated reflects a determination by FDA that an in vivo bioequivalence study is required.

(d) For certain drug products, bioavailability may be measured or bioequivalence may be demonstrated by evidence obtained in vitro in lieu of in vivo data. FDA shall waive the requirement for the submission of evidence obtained in vivo measuring the bioavailability or demonstrating the bioequivalence of the drug product if the drug product meets one of the following criteria:

(1) [Reserved]

(2) The drug product is in the same dosage form, but in a different strength, and is proportionally similar in its active and inactive ingredients to another drug product for which the same manufacturer has obtained approval and the conditions in paragraphs (d)(2)(i) through (d)(2)(iii) of this section are met:

(i) The bioavailability of this other drug product has been measured;

(ii) Both drug products meet an appropriate in vitro test approved by FDA; and

(iii) The applicant submits evidence showing that both drug products are proportionally similar in their active and inactive ingredients.

(iv) Paragraph (d) of this section does not apply to delayed release or extended release products.

(3) The drug product is, on the basis of scientific evidence submitted in the application, shown to meet an in vitro test that has been correlated with in vivo data.

(4) The drug product is a reformulated product that is identical, except for a different color, flavor, or preservative that could not affect the bioavailability of the reformulated product, to another drug product for which the same manufacturer has obtained approval and the following conditions are met:

(i) The bioavailability of the other product has been measured; and

(ii) Both drug products meet an appropriate in vitro test approved by FDA.

(e) FDA, for good cause, may waive a requirement for the submission of evidence of in vivo bioavailability or bioequivalence if waiver is compatible with the protection of the public health. For full new drug applications, FDA may defer a requirement for the submission of evidence of in vivo bioavailability if deferral is compatible with the protection of the public health.

(f) FDA, for good cause, may require evidence of in vivo bioavailability or bioequivalence for any drug product if the agency determines that any difference between the drug product and a listed drug may affect the bioavailability or bioequivalence of the drug product.

[57 FR 17998, Apr. 28, 1992, as amended at 67 FR 77673, Dec. 19, 2002]

§ 320.23 Basis for measuring in vivo bioavailability or demonstrating bioequivalence.

(a)(1) The in vivo bioavailability of a drug product is measured if the product's rate and extent of absorption, as determined by comparison of measured parameters, e.g., concentration of the active drug ingredient in the blood, urinary excretion rates, or pharmacological effects, do not indicate a significant difference from the reference material's rate and extent of absorption. For drug products that are not intended to be absorbed into the bloodstream, bioavailability may be assessed by measurements intended to reflect the rate and extent to which the active ingredient or active moiety becomes available at the site of action.

(2) Statistical techniques used shall be of sufficient sensitivity to detect

substance and, therefore, will not result in the introduction of any substance into the environment.

(f) Extramural contracts, other agreements, and grants for research for such purposes as to develop analytical methods or other test methodologies.

(g) Activities of voluntary Federal-State cooperative programs, including issuance of model regulations proposed for State adoption.

(h) Issuance, amendment, or revocation of procedural or administrative regulations and guidance documents, including procedures for submission of applications for product development, testing and investigational use, and approval.

(i) Corrections and technical changes in regulations.

(j) Issuance of CGMP regulations, HACCP regulations, establishment standards, emergency permit control regulations, GLP regulations, and issuance or denial of permits, exemptions, variances, or stays under these regulations.

(k) Establishment or repeal by regulation of labeling requirements for marketed articles if there will be no increase in the existing levels of use or change in the intended uses of the product or its substitutes.

(l) Routine maintenance and minor construction activities such as:

(1) Repair to or replacement of equipment or structural components (e.g., door, roof, or window) of facilities controlled by FDA;

(2) Lease extensions, renewals, or succeeding leases;

(3) Construction or lease construction of 10,000 square feet or less of occupiable space;

(4) Relocation of employees into existing owned or currently leased space;

(5) Acquisition of 20,000 square feet or less of occupiable space in a structure that was substantially completed before the issuance of solicitation for offers; and

(6) Acquisition of between 20,000 square feet and 40,000 square feet of occupiable space if it constitutes less than 40 percent of the occupiable space in a structure that was substantially completed before the solicitation for offers.

(m) Disposal of low-level radioactive waste materials (as defined in the Nuclear Regulatory Commission regulations at 10 CFR 61.2) and chemical waste materials generated in the laboratories serviced by the contracts administered by FDA, if the waste is disposed of in compliance with all applicable Federal, State, and local requirements.

[62 FR 40592, July 29, 1997, as amended at 65 FR 56479, Sept. 19, 2000]

§ 25.31 Human drugs and biologics.

The classes of actions listed in this section are categorically excluded and, therefore, ordinarily do not require the preparation of an EA or an EIS:

(a) Action on an NDA, abbreviated application, application for marketing approval of a biologic product, or a supplement to such applications, or action on an OTC monograph, if the action does not increase the use of the active moiety.

(b) Action on an NDA, abbreviated application, or a supplement to such applications, or action on an OTC monograph, if the action increases the use of the active moiety, but the estimated concentration of the substance at the point of entry into the aquatic environment will be below 1 part per billion.

(c) Action on an NDA, abbreviated application, application for marketing approval of a biologic product, or a supplement to such applications, or action on an OTC monograph, for substances that occur naturally in the environment when the action does not alter significantly the concentration or distribution of the substance, its metabolites, or degradation products in the environment.

(d) Withdrawal of approval of an NDA or an abbreviated application.

(e) Action on an IND.

(f) Testing and release by the Center for Biologics Evaluation and Research of lots or batches of a licensed biologic product.

(g) Establishment of bioequivalence requirements for a human drug or a comparability determination for a biologic product subject to licensing.

(h) Issuance, revocation, or amendment of a standard for a biologic product.

(i) Revocation of a license for a biologic product.

(j) Action on an application for marketing approval for marketing of a biologic product for transfusable human blood or blood components and plasma.

[62 FR 40592, July 29, 1997, as amended at 63 FR 26697, May 13, 1998; 64 FR 399, Jan. 5, 1999]

§ 25.32 Foods, food additives, and color additives.

The classes of actions listed in this section are categorically excluded and, therefore, ordinarily do not require the preparation of an EA or an EIS:

(a) Issuance, amendment, or repeal of a food standard.

(b) Action on a request for exemption for investigational use of a food additive if the food additive to be shipped under the request is intended to be used for clinical studies or research.

(c) Approval of a color additive petition to change a provisionally listed color additive to permanent listing for use in food, drugs, devices, or cosmetics.

(d) Testing and certification of batches of a color additive.

(e) Issuance of an interim food additive regulation.

(f) Affirmation of a food substance as GRAS for humans or animals on FDA's initiative or in response to a petition, under parts 182, 184, 186, or 582 of this chapter, and establishment or amendment of a regulation for a prior-sanctioned food ingredient, as defined in §§ 170.3(l) and 181.5(a) of this chapter, if the substance or food ingredient is already marketed in the United States for the proposed use.

(g) Issuance and enforcement of regulations relating to the control of communicable diseases or to interstate conveyance sanitation under parts 1240 and 1250 of this chapter.

(h) Approval of a request for diversion of adulterated or misbranded food for humans or animals to use as animal feeds.

(i) Approval of a food additive petition or GRAS affirmation petition, the granting of a request for exemption from regulation as a food additive under § 170.39 of this chapter, or allowing a notification submitted under 21 U.S.C. 348(h) to become effective, when the substance is present in finished food-packaging material at not greater than 5 percent-by-weight and is expected to remain with finished food-packaging material through use by consumers or when the substance is a component of a coating of a finished food-packaging material.

(j) Approval of a food additive petition or GRAS affirmation petition, the granting of a request for exemption from regulation as a food additive under § 170.39 of this chapter, or allowing a notification submitted under 21 U.S.C. 348(h) to become effective, when the substance is to be used as a component of a food-contact surface of permanent or semipermanent equipment or of another food-contact article intended for repeated use.

(k) Approval of a food additive petition, color additive petition, or GRAS affirmation petition, or allowing a notification submitted under 21 U.S.C. 348(h) to become effective, for substances added directly to food that are intended to remain in food through ingestion by consumers and that are not intended to replace macronutrients in food.

(l) Approval of a petition for color additives used in contact lenses, sutures, filaments used as supporting haptics in intraocular lenses, bone cement, and in other FDA-regulated products having similarly low levels of use.

(m) Action to prohibit or otherwise restrict or reduce the use of a substance in food, food packaging, or cosmetics.

(n) Issuance, amendment, or revocation of a regulation pertaining to infant formulas.

(o) Approval of a food additive petition for the intended expression product(s) present in food derived from new plant varieties.

(p) Issuance, amendment, or revocation of a regulation in response to a reference amount petition as described in § 101.12(h) of this chapter, a nutrient content claim petition as described in § 101.69 of this chapter, a health claim petition as described in § 101.70 of this chapter, or a petition pertaining to the label declaration of ingredients as described in § 101.103 of this chapter.

E

Guidance for Industry

Waiver of In Vivo Bioavailability and Bioequivalence Studies for Immediate-Release Solid Oral Dosage Forms Based on a Biopharmaceutics Classification System

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
August 2000
BP

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U.S. Department of Health and Human Services
Food and Drug Administration
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GUIDANCE FOR INDUSTRY[1]

Waiver of In Vivo Bioavailability and Bioequivalence Studies for Immediate-Release Solid Oral Dosage Forms Based on a Biopharmaceutics Classification System

I. INTRODUCTION

This guidance provides recommendations for sponsors of investigational new drug applications (INDs), new drug applications (NDAs), abbreviated new drug applications (ANDAs), and supplements to these applications who wish to request a waiver of in vivo bioavailability (BA) and/or bioequivalence (BE) studies for immediate release (IR) solid oral dosage forms. These waivers are intended to apply to (1) subsequent in vivo BA or BE studies of formulations after the initial establishment of the in vivo BA of IR dosage forms during the IND period, and (2) in vivo BE studies of IR dosage forms in ANDAs. Regulations at 21 CFR part 320 address the requirements for bioavailability (BA) and BE data for approval of drug applications and supplemental applications. Provision for waivers of in vivo BA/BE studies (biowaivers) under certain conditions is provided at 21 CFR 320.22. This guidance explains when biowaivers can be requested for IR solid oral dosage forms based on an approach termed the Biopharmaceutics Classification System (BCS).

II. THE BIOPHARMACEUTICS CLASSIFICATION SYSTEM

The BCS is a scientific framework for classifying drug substances based on their aqueous solubility and intestinal permeability. When combined with the dissolution of the drug product, the BCS takes into account three major factors that govern the rate and extent of drug absorption from IR solid oral dosage forms: dissolution, solubility, and intestinal permeability.[2] According to the BCS, drug substances are classified as follows:

Class 1:	High Solubility – High Permeability
Class 2:	Low Solubility – High Permeability
Class 3:	High Solubility – Low Permeability

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[1] This guidance has been prepared by the Biopharmaceutics Classification System Working Group of the Biopharmaceutics Coordinating Committee in the Center for Drug Evaluation and Research (CDER) at the Food and Drug Administration (FDA). This guidance represents the Agency's current thinking on the topic. It does not create or confer any rights for or on any person and does not operate to bind FDA or the public. An alternative approach may be used if such an approach satisfies the requirements of the applicable statutes, regulations, or both.

[2] Amidon, G. L., H. Lennernäs, V. P. Shah, and J. R. Crison, "A Theoretical Basis For a Biopharmaceutics Drug Classification: The Correlation of In Vitro Drug Product Dissolution and In Vivo Bioavailability," *Pharmaceutical Research*, 12: 413-420 (1995).

Class 4: Low Solubility – Low Permeability

In addition, IR solid oral dosage forms are categorized as having rapid or slow dissolution. Within this framework, when certain criteria are met, the BCS can be used as a drug development tool to help sponsors justify requests for biowaivers.

Observed in vivo differences in the rate and extent of absorption of a drug from two pharmaceutically equivalent solid oral products may be due to differences in drug dissolution in vivo.[2] However, when the in vivo dissolution of an IR solid oral dosage form is rapid in relation to gastric emptying and the drug has high permeability, the rate and extent of drug absorption is unlikely to be dependent on drug dissolution and/or gastrointestinal transit time. Under such circumstances, demonstration of in vivo BA or BE may not be necessary for drug products containing Class 1 drug substances, as long as the inactive ingredients used in the dosage form do not significantly affect absorption of the active ingredients. The BCS approach outlined in this guidance can be used to justify biowaivers for *highly soluble* and *highly permeable* drug substances (i.e., Class 1) in IR solid oral dosage forms that exhibit *rapid in vitro dissolution* using the recommended test methods (21 CFR 320.22(e)). The recommended methods for determining solubility, permeability, and in vitro dissolution are discussed below.

A. Solubility

The solubility class boundary is based on the highest dose strength of an IR product that is the subject of a biowaiver request. A drug substance is considered *highly soluble* when the highest dose strength is soluble in 250 ml or less of aqueous media over the pH range of 1-7.5. The volume estimate of 250 ml is derived from typical BE study protocols that prescribe administration of a drug product to fasting human volunteers with a glass (about 8 ounces) of water.

B. Permeability

The permeability class boundary is based indirectly on the extent of absorption (fraction of dose absorbed, not systemic BA) of a drug substance in humans and directly on measurements of the rate of mass transfer across human intestinal membrane. Alternatively, nonhuman systems capable of predicting the extent of drug absorption in humans can be used (e.g., in vitro epithelial cell culture methods). In the absence of evidence suggesting instability in the gastrointestinal tract, a drug substance is considered to be *highly permeable* when the extent of absorption in humans is determined to be 90% or more of an administered dose based on a mass balance determination or in comparison to an intravenous reference dose.

C. Dissolution

In this guidance, an IR drug product is considered *rapidly dissolving* when no less than 85% of the labeled amount of the drug substance dissolves within 30

minutes, using *U.S. Pharmacopeia* (USP) Apparatus I at 100 rpm (or Apparatus II at 50 rpm) in a volume of 900 ml or less in each of the following media: (1) 0.1 N HCl or Simulated Gastric Fluid USP without enzymes; (2) a pH 4.5 buffer; and (3) a pH 6.8 buffer or Simulated Intestinal Fluid USP without enzymes.

III. METHODOLOGY FOR CLASSIFYING A DRUG SUBSTANCE AND FOR DETERMINING THE DISSOLUTION CHARACTERISTICS OF A DRUG PRODUCT

The following approaches are recommended for classifying a drug substance and determining the dissolution characteristics of an IR drug product according to the BCS:

A. Determining Drug Substance Solubility Class

An objective of the BCS approach is to determine the equilibrium solubility of a drug substance under physiological pH conditions. The pH-solubility profile of the test drug substance should be determined at 37 ± 1°C in aqueous media with a pH in the range of 1-7.5. A sufficient number of pH conditions should be evaluated to accurately define the pH-solubility profile. The number of pH conditions for a solubility determination can be based on the ionization characteristics of the test drug substance. For example, when the pKa of a drug is in the range of 3-5, solubility should be determined at pH = pKa, pH = pKa +1, pH = pKa-1, and at pH = 1 and 7.5. A minimum of three replicate determinations of solubility in each pH condition is recommended. Depending on study variability, additional replication may be necessary to provide a reliable estimate of solubility. Standard buffer solutions described in the USP are considered appropriate for use in solubility studies. If these buffers are not suitable for physical or chemical reasons, other buffer solutions can be used. Solution pH should be verified after addition of the drug substance to a buffer. Methods other than the traditional shake-flask method, such as acid or base titration methods, can also be used with justification to support the ability of such methods to predict equilibrium solubility of the test drug substance. Concentration of the drug substance in selected buffers (or pH conditions) should be determined using a validated stability-indicating assay that can distinguish the drug substance from its degradation products.[3] If degradation of the drug substance is observed as a function of buffer composition and/or pH, it should be reported along with other stability data recommended in section III.B.3.

The solubility class should be determined by calculating the volume of an aqueous medium sufficient to dissolve the highest dose strength in the pH range of 1-7.5. A drug substance should be classified as highly soluble when the highest dose strength is soluble in ≤ 250 ml of aqueous media over the pH range of 1-7.5.

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[3] See the FDA guidance for industry on *Submitting Documentation for the Stability of Human Drugs and Biologics* (February 1987), posted at http://www.fda.gov/guidance/index.htm.

B. Determining Drug Substance Permeability Class

The permeability class of a drug substance can be determined in human subjects using mass balance, absolute BA, or intestinal perfusion approaches. Recommended methods not involving human subjects include in vivo or in situ intestinal perfusion in a suitable animal model (e.g., rats), and/or in vitro permeability methods using excised intestinal tissues, or monolayers of suitable epithelial cells. In many cases, a single method may be sufficient (e.g., when the absolute BA is 90% or more, or when 90% or more of the administered drug is recovered in urine). When a single method fails to conclusively demonstrate a permeability classification, two different methods may be advisable. Chemical structure and/or certain physicochemical attributes of a drug substance (e.g., partition coefficient in suitable systems) can provide useful information about its permeability characteristics. Sponsors may wish to consider use of such information to further support a classification.

1. Pharmacokinetic Studies in Humans

a. Mass Balance Studies

Pharmacokinetic mass balance studies using unlabeled, stable isotopes or a radiolabeled drug substance can be used to document the extent of absorption of a drug. Depending on the variability of the studies, a sufficient number of subjects should be enrolled to provide a reliable estimate of extent of absorption. Because this method can provide highly variable estimates of drug absorption for many drugs, other methods described below may be preferable.

b. Absolute Bioavailability Studies

Oral BA determination using intravenous administration as a reference can be used. Depending on the variability of the studies, a sufficient number of subjects should be enrolled in a study to provide a reliable estimate of the extent of absorption. When the absolute BA of a drug is shown to be 90% or more, additional data to document drug stability in the gastrointestinal fluid is not necessary.

2. Intestinal Permeability Methods

The following methods can be used to determine the permeability of a drug substance from the gastrointestinal tract: (1) in vivo intestinal perfusion studies in humans; (2) in vivo or in situ intestinal perfusion studies using suitable animal models; (3) in vitro permeation studies using excised human or animal intestinal tissues; or (4) in vitro permeation studies across a monolayer of cultured epithelial cells.

In vivo or in situ animal models and in vitro methods, such as those using cultured monolayers of animal or human epithelial cells, are considered appropriate for passively transported drugs. The observed low permeability of some drug substances in humans could be caused by efflux of drugs via membrane transporters such as P-glycoprotein (P-gp). When the efflux transporters are absent in these models, or their degree of expression is low compared to that in humans, there may be a greater likelihood of misclassification of permeability class for a drug subject to efflux compared to a drug transported passively. Expression of known transporters in selected study systems should be characterized. Functional expression of efflux systems (e.g., P-gp) can be demonstrated with techniques such as bidirectional transport studies, demonstrating a higher rate of transport in the basolateral-to-apical direction as compared to apical-to-basolateral direction using selected model drugs or chemicals at concentrations that do not saturate the efflux system (e.g., cyclosporin A, vinblastine, rhodamine 123). An acceptance criterion for intestinal efflux that should be present in a test system cannot be set at this time. Instead, this guidance recommends limiting the use of nonhuman permeability test methods for drug substances that are transported by passive mechanisms. Pharmacokinetic studies on dose linearity or proportionality may provide useful information for evaluating the relevance of observed in vitro efflux of a drug. For example, there may be fewer concerns associated with the use of in vitro methods for a drug that has a higher rate of transport in the basolateral-to-apical direction at low drug concentrations but exhibits linear pharmacokinetics in humans.

For application of the BCS, an apparent passive transport mechanism can be assumed when one of the following conditions is satisfied:

- A linear (pharmacokinetic) relationship between the dose (e.g., relevant clinical dose range) and measures of BA (area under the concentration-time curve) of a drug is demonstrated in humans

- Lack of dependence of the measured in vivo or in situ permeability is demonstrated in an animal model on initial drug concentration (e.g., 0.01, 0.1, and 1 times the highest dose strength dissolved in 250 ml) in the perfusion fluid

- Lack of dependence of the measured in vitro permeability on initial drug concentration (e.g., 0.01, 0.1, and 1 times the highest dose strength dissolved in 250 ml) is demonstrated in donor fluid and transport direction (e.g., no statistically significant difference in the rate of transport between the apical-to-basolateral and basolateral-to-apical direction for the drug concentrations selected) using a suitable in vitro cell culture method that has been shown to express known efflux transporters (e.g., P-gp)

To demonstrate suitability of a permeability method intended for application of the BCS, a rank-order relationship between test permeability values and the extent of drug absorption data in human subjects should be established using a sufficient number of model drugs. For in vivo intestinal perfusion studies in humans, six model drugs are recommended. For in vivo or in situ intestinal perfusion studies in animals and for in vitro cell culture methods, twenty model drugs are recommended. Depending on study variability, a sufficient number of subjects, animals, excised tissue samples, or cell monolayers should be used in a study to provide a reliable estimate of drug permeability. This relationship should allow precise differentiation between drug substances of low and high intestinal permeability attributes.

For demonstration of suitability of a method, model drugs should represent a range of low (e.g., < 50%), moderate (e.g., 50 - 89%), and high (≥ 90%) absorption. Sponsors may select compounds from the list of drugs and/or chemicals provided in Attachment A or they may choose to select other drugs for which there is information available on mechanism of absorption and reliable estimates of the extent of drug absorption in humans.

After demonstrating suitability of a method and maintaining the same study protocol, it is not necessary to retest all selected model drugs for subsequent studies intended to classify a drug substance. Instead, a low and a high permeability model drug should be used as internal standards (i.e., included in the perfusion fluid or donor fluid along with the test drug substance). These two internal standards are in addition to the fluid volume marker (or a zero permeability compound such as PEG 4000) that is included in certain types of perfusion techniques (e.g., closed loop techniques). The choice of internal standards should be based on compatibility with the test drug substance (i.e., they should not exhibit any significant physical, chemical, or permeation interactions). When it is not feasible to follow this protocol, the permeability of internal standards should be determined in the same subjects, animals, tissues, or monolayers, following evaluation of the test drug substance. The permeability values of the two internal standards should not differ significantly between different tests, including those conducted to demonstrate suitability of the method. At the end of an in situ or in vitro test, the amount of drug in the membrane should be determined.

For a given test method with set conditions, selection of a high permeability internal standard with permeability in close proximity to the low/high permeability class boundary may facilitate classification of a test drug substance. For instance, a test drug substance may be determined to be highly permeable when its permeability value is equal to or greater than that of the selected internal standard with high permeability.

3. Instability in the Gastrointestinal Tract

Determining the extent of absorption in humans based on mass balance studies using total radioactivity in urine does not take into consideration the extent of degradation of a drug in the gastrointestinal fluid prior to intestinal membrane permeation. In addition, some methods for determining permeability could be based on loss or clearance of a drug from fluids perfused into the human and/or animal gastrointestinal tract either in vivo or in situ. Documenting the fact that drug loss from the gastrointestinal tract arises from intestinal membrane permeation, rather than a degradation process, will help establish permeability. Stability in the gastrointestinal tract may be documented using gastric and intestinal fluids obtained from human subjects. Drug solutions in these fluids should be incubated at 37°C for a period that is representative of in vivo drug contact with these fluids; for example, 1 hour in gastric fluid and 3 hours in intestinal fluid. Drug concentrations should then be determined using a validated stability-indicating assay method. Significant degradation (>5%) of a drug in this protocol could suggest potential instability. Obtaining gastrointestinal fluids from human subjects requires intubation and may be difficult in some cases. Use of gastrointestinal fluids from suitable animal models and/or simulated fluids such as Gastric and Intestinal Fluids USP can be substituted when properly justified.

C. Determining Drug Product Dissolution Characteristics and Dissolution Profile Similarity[4]

Dissolution testing should be carried out in USP Apparatus I at 100 rpm or Apparatus II at 50 rpm using 900 ml of the following dissolution media: (1) 0.1 N HCl or Simulated Gastric Fluid USP without enzymes; (2) a pH 4.5 buffer; and (3) a pH 6.8 buffer or Simulated Intestinal Fluid USP without enzymes. For capsules and tablets with gelatin coating, Simulated Gastric and Intestinal Fluids USP (with enzymes) can be used.

Dissolution testing apparatus used in this evaluation should conform to the requirements in USP (<711> Dissolution). Selection of the dissolution testing apparatus (USP Apparatus I or II) during drug development should be based on a comparison of in vitro dissolution and in vivo pharmacokinetic data available for the product. The USP Apparatus I (*basket method*) is generally preferred for capsules and products that tend to float, and USP Apparatus II (*paddle method*) is generally preferred for tablets. For some tablet dosage forms, in vitro (but not in vivo) dissolution may be slow due to the manner in which the disintegrated product settles at the bottom of a dissolution vessel. In such situations, USP Apparatus I may be preferred over Apparatus II. If the testing conditions need to be modified to better reflect rapid in vivo dissolution (e.g., use of a different rotating speed), such modifications can be justified by comparing in vitro

[4] See the FDA guidance for industry on *Dissolution Testing of Immediate Release Solid Oral Dosage Forms* (August 1997).

dissolution with in vivo absorption data (e.g., a relative BA study using a simple aqueous solution as the reference product).
A minimum of 12 dosage units of a drug product should be evaluated to support a biowaiver request. Samples should be collected at a sufficient number of intervals to characterize the dissolution profile of the drug product (e.g., 10, 15, 20, and 30 minutes).

When comparing the test and reference products, dissolution profiles should be compared using a similarity factor (f_2). The similarity factor is a logarithmic reciprocal square root transformation of the sum of squared error and is a measurement of the similarity in the percent (%) of dissolution between the two curves.

$$f_2 = 50 \bullet \log \{[1+(1/n)\bullet \sum_{t=1}^{n} (R_t - T_t)^2]^{-0.5} \bullet 100\}$$

Two dissolution profiles are considered similar when the f_2 value is • 50. To allow the use of mean data, the coefficient of variation should not be more than 20% at the earlier time points (e.g., 10 minutes), and should not be more than 10% at other time points. Note that when both test and reference products dissolve 85% or more of the label amount of the drug in • 15 minutes using all three dissolution media recommended above, the profile comparison with an f_2 test is unnecessary.

IV. ADDITIONAL CONSIDERATIONS FOR REQUESTING A BIOWAIVER

When requesting a BCS-based waiver for in vivo BA/BE studies for IR solid oral dosage forms, applicants should note that the following factors can affect their request or the documentation of their request:

A. Excipients

Excipients can sometimes affect the rate and extent of drug absorption. In general, using excipients that are currently in FDA-approved IR solid oral dosage forms will not affect the rate or extent of absorption of a highly soluble and highly permeable drug substance that is formulated in a rapidly dissolving IR product. To support a biowaiver request, the quantity of excipients in the IR drug product should be consistent with the intended function (e.g., lubricant). When new excipients or atypically large amounts of commonly used excipients are included in an IR solid dosage form, additional information documenting the absence of an impact on BA of the drug may be requested by the Agency. Such information can be provided with a relative BA study using a simple aqueous solution as the reference product. Large quantities of certain excipients, such as surfactants (e.g., polysorbate 80) and sweeteners (e.g., mannitol or sorbitol) may be problematic, and sponsors are encouraged to contact the review division when this is a factor.

B. Prodrugs

Permeability of prodrugs will depend on the mechanism and (anatomical) site of conversion to the drug substance. When the prodrug-to-drug conversion is shown to occur predominantly after intestinal membrane permeation, the permeability of the prodrug should be measured. When this conversion occurs prior to intestinal permeation, the permeability of the drug should be determined. Dissolution and pH-solubility data on both prodrug and drug can be relevant. Sponsors may wish to consult with appropriate review staff before applying the BCS approach to IR products containing prodrugs.

C. Exceptions

BCS-based biowaivers are not applicable for the following:

1. Narrow Therapeutic Range Drugs[5]

This guidance defines narrow therapeutic range drug products as those containing certain drug substances that are subject to therapeutic drug concentration or pharmacodynamic monitoring, and/or where product labeling indicates a narrow therapeutic range designation. Examples include digoxin, lithium, phenytoin, theophylline, and warfarin. Because not all drugs subject to therapeutic drug concentration or pharmacodynamic monitoring are narrow therapeutic range drugs, sponsors should contact the appropriate review division to determine whether a drug should be considered to have a narrow therapeutic range.

2. Products Designed to be Absorbed in the Oral Cavity

A request for a waiver of in vivo BA/BE studies based on the BCS is not appropriate for dosage forms intended for absorption in the oral cavity (e.g., sublingual or buccal tablets).

V. REGULATORY APPLICATIONS OF THE BCS

A. INDs/NDAs

Evidence demonstrating in vivo BA or information to permit FDA to waive this evidence must be included in NDAs (21 CFR 320.21(a)). A specific objective is to establish in vivo performance of the dosage form used in the clinical studies that provided primary evidence of efficacy and safety. The sponsor may wish to determine the relative BA of an IR solid oral dosage form by comparison with an oral solution, suspension, or intravenous injection (21 CFR 320.25 (d)(2) and

[5] This guidance uses the term *narrow therapeutic range* instead of *narrow therapeutic index*, although the latter is more commonly used.

320.25 (d)(3)). The BA of the clinical trial dosage form should be optimized during the IND period.

Once the in vivo BA of a formulation is established during the IND period, waivers of subsequent in vivo BE studies, following major changes in components, composition, and/or method of manufacture (e.g., similar to SUPAC-IR Level 3 changes[6]) may be possible using the BCS. BCS-based biowaivers are applicable to the to-be-marketed formulation when changes in components, composition, and/or method of manufacture occur to the clinical trial formulation, as long as the dosage forms have rapid and similar in vitro dissolution profiles (see sections II and III). This approach is useful only when the drug substance is highly soluble and highly permeable (BCS Class 1), and the formulations pre- and postchange are *pharmaceutical equivalents* (under the definition at 21 CFR 320.1 (c)). BCS-based biowaivers are intended only for BE studies. They do not apply to food effect BA studies or other pharmacokinetic studies.

B. ANDAs

BCS-based biowaivers can be requested for rapidly dissolving IR test products containing highly soluble and highly permeable drug substances, provided that the reference listed drug product is also rapidly dissolving and the test product exhibits similar dissolution profiles to the reference listed drug product (see sections II and III). This approach is useful when the test and reference dosage forms are pharmaceutical equivalents. The choice of dissolution apparatus (USP Apparatus I or II) should be the same as that established for the reference listed drug product.

C. Postapproval Changes

BCS-based biowaivers can be requested for significant postapproval changes (e.g., Level 3 changes in components and composition) to a rapidly dissolving IR product containing a highly soluble, highly permeable drug substance, provided that dissolution remains rapid for the postchange product and both pre- and postchange products exhibit similar dissolution profiles (see sections II and III). This approach is useful only when the drug products pre- and postchange are pharmaceutical equivalents.

VI. DATA TO SUPPORT A REQUEST FOR BIOWAIVERS

The drug substance for which a waiver is being requested should be highly soluble and highly permeable. Sponsors requesting biowaivers based on the BCS should submit the following information to the Agency for review by the Office of Clinical Pharmacology

[6] See the FDA guidance for industry on *Immediate Release Solid Oral Dosage Forms: Scale-Up and Post-Approval Changes* (November 1995).

and Biopharmaceutics (for NDAs) or Office of Generic Drugs, Division of Bioequivalence (for ANDAs):

A. Data Supporting High Solubility

Data supporting high solubility of the test drug substance should be developed (see section III.A). The following information should be included in the application:

- A description of test methods, including information on analytical method and composition of the buffer solutions
- Information on chemical structure, molecular weight, nature of the drug substance (acid, base, amphoteric, or neutral), and dissociation constants (pKa(s))
- Test results (mean, standard deviation, and coefficient of variation) summarized in a table under solution pH, drug solubility (e.g., mg/ml), and volume of media required to dissolve the highest dose strength
- A graphic representation of mean pH-solubility profile

B. Data Supporting High Permeability

Data supporting high permeability of the test drug substance should be developed (see section III.B). The following information should be included in the application:

- For human pharmacokinetic studies, information on study design and methods used along with the pharmacokinetic data
- For direct permeability methods, information supporting the suitability of a selected method that encompasses a description of the study method, criteria for selection of human subjects, animals, or epithelial cell line, drug concentrations in the donor fluid, description of the analytical method, method used to calculate extent of absorption or permeability, and where appropriate, information on efflux potential (e.g., bidirectional transport data)
- A list of selected model drugs along with data on extent of absorption in humans (mean, standard deviation, coefficient of variation) used to establish suitability of a method, permeability values for each model drug (mean, standard deviation, coefficient of variation), permeability class of each model drug, and a plot of the extent of absorption as a function of permeability (mean ± standard deviation or 95% confidence interval) with identification of the low/high permeability

class boundary and selected internal standard. Information to support high permeability of a test drug substance should include permeability data on the test drug substance, the internal standards (mean, standard deviation, coefficient of variation), stability information, data supporting passive transport mechanism where appropriate, and methods used to establish high permeability of the test drug substance.

C. Data Supporting Rapid and Similar Dissolution

For submission of a biowaiver request, an IR product should be rapidly dissolving. Data supporting rapid dissolution attributes of the test and reference products should be developed (see section III.C). The following information should be included in the application:

- A brief description of the IR products used for dissolution testing, including information on batch or lot number, expiry date, dimensions, strength, and weight

- Dissolution data obtained with 12 individual units of the test and reference products using recommended test methods in section III.C. The percentage of labeled claim dissolved at each specified testing interval should be reported for each individual dosage unit. The mean percent dissolved, range (highest and lowest) of dissolution, and coefficient of variation (relative standard deviation) should be tabulated. A graphic representation of the mean dissolution profiles for the test and reference products in the three media should also be included.

- Data supporting similarity in dissolution profiles between the test and reference products in each of the three media, using the f_2 metric

D. Additional Information

The manufacturing process used to make the test product should be described briefly to provide information on the method of manufacture (e.g., wet granulation vs. direct compression). A list of excipients used, the amount used, and their intended functions should be provided. Excipients used in the test product should have been used previously in FDA-approved IR solid oral dosage forms.

ATTACHMENT A

This attachment includes model drugs suggested for use in establishing suitability of a permeability method as described in section III. The permeability of these compounds was determined based on data available to the FDA. Potential *internal standards* (IS) and *efflux pump substrates* (ES) are also identified.

Drug	Permeability Class
Antipyrine	High (Potential IS candidate)
Caffeine	High
Carbamazepine	High
Fluvastatin	High
Ketoprofen	High
Metoprolol	High (Potential IS candidate)
Naproxen	High
Propranolol	High
Theophylline	High
Verapamil	High (Potential ES candidate)
Amoxicillin	Low
Atenolol	Low
Furosemide	Low
Hydrochlorthiazide	Low
Mannitol	Low (Potential IS candidate)
• -Methyldopa	Low
Polyethylene glycol (400)	Low
Polyethylene glycol (1000)	Low
Polyethylene glycol (4000)	Low (Zero permeability marker)
Ranitidine	Low

F

FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH
APPROVED DRUG PRODUCTS
with
Therapeutic Equivalence Evaluations

24th Edition

PREFACE

The publication, ***Approved Drug Products with Therapeutic Equivalence Evaluations*** (the List), identifies drug products approved on the basis of safety and effectiveness by the Food and Drug Administration (FDA) under the Federal Food, Drug, and Cosmetic Act (the Act). Drugs on the market approved only on the basis of safety (covered by the ongoing Drug Efficacy Study Implementation [DESI] review [e.g., Donnatal® Tablets and Librax® Capsules] or pre-1938 drugs [e.g., Phenobarbital Tablets]) are not included in this publication. The main criterion for the inclusion of any product is that the product is the subject of an application with an effective approval that has not been withdrawn for safety or efficacy reasons. Inclusion of products on the List is independent of any current regulatory action through administrative or judicial means against a drug product. In addition, the List contains therapeutic equivalence evaluations for approved multisource prescription drug products. These evaluations have been prepared to serve as public information and advice to state health agencies, prescribers, and pharmacists to promote public education in the area of drug product selection and to foster containment of health care costs. Therapeutic equivalence evaluations in this publication are not official FDA actions affecting the legal status of products under the Act.

Background of the Publication. To contain drug costs, virtually every state has adopted laws and/or regulations that encourage the substitution of drug products. These state laws generally require either that substitution be limited to drugs on a specific list (the positive formulary approach) or that it be permitted for all drugs except those prohibited by a particular list (the negative formulary approach). Because of the number of requests in the late 1970s for FDA assistance in preparing both positive and negative formularies, it became apparent that FDA could not serve the needs of each state on an individual basis. The Agency also recognized that providing a single list based on common criteria would be preferable to evaluating drug products on the basis of differing definitions and criteria in various state laws. As a result, on May 31, 1978, the Commissioner of the Food and Drug Administration sent a letter to officials of each state stating FDA's intent to provide a list of all prescription drug products that are approved by

FDA for safety and effectiveness, along with therapeutic equivalence determinations for multisource prescription products.

The List was distributed as a proposal in January 1979. It included only currently marketed prescription drug products approved by FDA through new drug applications (NDAs) and abbreviated new drug applications (ANDAs) under the provisions of Section 505 of the Act.

The therapeutic equivalence evaluations in the List reflect FDA's application of specific criteria to the approved multisource prescription drug products on the List. These evaluations are presented in the form of code letters that indicate the basis for the evaluation made. An explanation of the code appears in the *Introduction*.

A complete discussion of the background and basis of FDA's therapeutic equivalence evaluation policy was published in the *Federal Register* on January 12, 1979 (44 FR 2932). The final rule, which includes FDA's responses to the public comments on the proposal, was published in the *Federal Register* on October 31, 1980 (45 FR 72582). The first publication, October 1980, of the final version of the List incorporated appropriate corrections and additions. Each subsequent edition has included the new approvals and made appropriate changes in data.

On September 24, 1984, the President signed into law the Drug Price Competition and Patent Term Restoration Act (1984 Amendments). The 1984 Amendments require that FDA, among other things, make publicly available a list of approved drug products that is updated monthly. The ***Approved Drug Products with Therapeutic Equivalence Evaluations*** publication satisfies this requirement. The *Addendum* to this publication identifies drugs that qualify under the 1984 Amendments for periods of exclusivity (during which ANDAs or applications described in Section 505(b)(2) of the Act for those drugs may not be submitted for a specified period of time and, if allowed to be submitted, would be tentatively approved) and provides patent information concerning the listed drugs which also may delay the approval of ANDAs or Section 505(b)(2) applications. The *Addendum* also provides additional information that may be helpful to those submitting a new drug application to the Agency.

The Agency intends to use this publication to further its objective of obtaining input and comment on the publication itself and related Agency procedures. Therefore, if you have comments on how the publication can be improved, please send them to the Director, Division of Labeling and Program Support HFD-610, Office of Generic Drugs, Center for Drug and Evaluation and Research, 7500 Standish Place, Rockville, MD 20855. Comments received are publicly available to the extent allowable under the Freedom of Information regulations.

INTRODUCTION

Content and Exclusion

The List is composed of four parts: (1) approved prescription drug products with therapeutic equivalence evaluations; (2) approved over-the-counter (OTC) drug products for those drugs that may not be marketed without NDAs or ANDAs because they are not covered under existing OTC monographs; (3) drug products with approval under Section 505 of the Act administered by the

Center for Biologics Evaluation and Research; and (4) a cumulative list of approved products that have never been marketed, are for exportation, are for military use, have been discontinued from marketing, or have had their approvals withdrawn for other than safety or efficacy reasons subsequent to being discontinued from marketing. All established names for active ingredients generally conform to official compendial names or *United States Adopted Names* (USAN) as prescribed in (21 CFR 299.4(e)). In addition, a list of uniform terms is provided. An *Addendum* contains drug patent and exclusivity information for the Prescription and OTC Drug Product Lists, and for the Drug Products with Approval under Section 505 of the Act Administered by the Center for Biologics Evaluation and Research.

Prior to the 6th Edition, the publication had excluded OTC drug products and drug products with approval under Section 505 of the Act Administered by the Center for Biologics Evaluation and Research because the main purpose of the publication was to provide information to states regarding FDA's recommendation as to which generic prescription drug products were acceptable candidates for drug product selection. The 1984 Amendments required the Agency to begin publishing an up-to-date list of all marketed drug products, OTC as well as prescription, that have been approved for safety and efficacy and for which new drug applications are required.

Under the 1984 Amendments, some drug products were given tentative approvals. Prior to the effective date, the Agency will not include drug products with tentative approval in the List; however, they are available in the *FDA Drug Product Approvals List* on the Internet World Wide Web. When the tentative approval becomes a full approval through a subsequent action letter to the application holder, the Agency will list the drug product and the final, effective approval date in the appropriate approved drug product list.

Distributors or repackagers of products on the List are not identified. Because distributors or repackagers are not required to notify FDA when they shift their sources of supply from one approved manufacturer to another, it is not possible to maintain complete information linking product approval with the distributor or repackager handling the products.

Therapeutic Equivalence-Related Terms

Pharmaceutical Equivalents. Drug products are considered pharmaceutical equivalents if they contain the same active ingredient(s), are of the same dosage form, route of administration and are identical in strength or concentration (e.g., chlordiazepoxide hydrochloride, 5mg capsules). Pharmaceutically equivalent drug products are formulated to contain the same amount of active ingredient in the same dosage form and to meet the same or compendial or other applicable standards (i.e., strength, quality, purity, and identity), but they may differ in characteristics such as shape, scoring configuration, release mechanisms, packaging, excipients (including colors, flavors, preservatives), expiration time, and, within certain limits, labeling.

Pharmaceutical Alternatives. Drug products are considered pharmaceutical alternatives if they contain the same therapeutic moiety, but are different salts, esters, or complexes of that moiety, or are different dosage forms or strengths (e.g., tetracycline hydrochloride, 250mg capsules vs. tetracycline phosphate complex, 250mg capsules; quinidine sulfate, 200mg tablets vs. quinidine sulfate, 200mg capsules). Data are generally not available for FDA to make the determination of tablet to capsule bioequivalence. Different dosage forms and strengths within a product line by a single manufacturer are thus pharmaceutical alternatives, as are extended-release products when compared with immediate- or standard-release formulations of the same

active ingredient.

Therapeutic Equivalents. Drug products are considered to be therapeutic equivalents only if they are pharmaceutical equivalents and if they can be expected to have the same clinical effect and safety profile when administered to patients under the conditions specified in the labeling.

FDA classifies as therapeutically equivalent those products that meet the following general criteria: (1) they are approved as safe and effective; (2) they are pharmaceutical equivalents in that they (a) contain identical amounts of the same active drug ingredient in the same dosage form and route of administration, and (b) meet compendial or other applicable standards of strength, quality, purity, and identity; (3) they are bioequivalent in that (a) they do not present a known or potential bioequivalence problem, and they meet an acceptable *in vitro* standard, or (b) if they do present such a known or potential problem, they are shown to meet an appropriate bioequivalence standard; (4) they are adequately labeled; and (5) they are manufactured in compliance with Current Good Manufacturing Practice regulations. *The concept of therapeutic equivalence, as used to develop the List, applies only to drug products containing the same active ingredient(s) and does not encompass a comparison of different therapeutic agents used for the same condition (e.g., propoxyphene hydrochloride vs. pentazocine hydrochloride for the treatment of pain).* Any drug product in the List repackaged and/or distributed by other than the application holder is considered to be therapeutically equivalent to the application holder's drug product even if the application holder's drug product is single source or coded as non-equivalent (e.g., **BN**). Also, distributors or repackagers of an application holder's drug product are considered to have the same code as the application holder. Therapeutic equivalence determinations are not made for unapproved, off-label indications.

FDA considers drug products to be therapeutically equivalent if they meet the criteria outlined above, even though they may differ in certain other characteristics such as shape, scoring configuration, release mechanisms, packaging, excipients (including colors, flavors, preservatives), expiration date/time and minor aspects of labeling (e.g., the presence of specific pharmacokinetic information) and storage conditions. When such differences are important in the care of a particular patient, it may be appropriate for the prescribing physician to require that a particular brand be dispensed as a medical necessity. With this limitation, however, FDA believes that products classified as therapeutically equivalent can be substituted with the full expectation that the substituted product will produce the same clinical effect and safety profile as the prescribed product.

Bioavailability. This term means the rate and extent to which the active ingredient or active moiety is absorbed from a drug product and becomes available at the site of action. For drug products that are not intended to be absorbed into the bloodstream, bioavailability may be assessed by measurements intended to reflect the rate and extent to which the active ingredient or active moiety becomes available at the site of action.

Bioequivalent Drug Products. This term describes pharmaceutical equivalent or alternative products that display comparable bioavailability when studied under similar experimental conditions. Section 505 (j)(7)(B) of the Act describes one set of conditions under which a test and reference listed drug shall be considered bioequivalent:

> the rate and extent of absorption of the test drug do not show a significant difference from the rate and extent of absorption of the

reference drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses; or

the extent of absorption of the test drug does not show a significant difference from the extent of absorption of the reference drug when administered at the same molar dose of the therapeutic ingredient under similar experimental conditions in either a single dose or multiple doses and the difference from the reference drug in the rate of absorption of the drug is intentional, is reflected in its proposed labeling, is not essential to the attainment of effective body drug concentrations on chronic use, and is considered medically insignificant for the drug.

Where these above methods are not applicable (e.g., for drug products that are not intended to be absorbed into the bloodstream), other *in vivo* or *in vitro* test methods to demonstrate bioequivalence may be appropriate.

Bioequivalence may sometimes be demonstrated using an *in vitro* bioequivalence standard, especially when such an *in vitro* test has been correlated with human *in vivo* bioavailability data. In other situations, bioequivalence may sometimes be demonstrated through comparative clinical trials or pharmacodynamic studies.

Statistical Criteria for Bioequivalence

Under the Drug Price Competition and Patent Term Restoration Act of 1984, manufacturers seeking approval to market a generic drug product must submit data demonstrating that the drug product is bioequivalent to the pioneer (innovator) drug product. A major premise underlying the 1984 law is that bioequivalent drug products are therapeutically equivalent and, therefore, interchangeable.

Bioavailability refers to the rate and extent to which the active ingredient or therapeutic ingredient is absorbed from a drug product and becomes available at the site of drug action (Federal Food, Drug and Cosmetic Act, section 505(j)(8)). Bioequivalence refers to equivalent release of the same drug substance from two or more drug products or formulations. This leads to an equivalent rate and extent of absorption from these formulations. Underlying the concept of bioequivalence is the thesis that, if a drug product contains a drug substance that is chemically identical and is delivered to the site of action at the same rate and extent as another drug product, then it is equivalent and can be substituted for that drug product. Methods used to define bioequivalence can be found in 21 CFR 320.24, and include (1) pharmacokinetic (PK) studies, (2) pharmacodynamic (PD) studies, (3) comparative clinical trials, and (4) in-vitro studies. The choice of study used is based on the site of action of the drug and the ability of the study design to compare drug delivered to that site by the two products.

The standard bioequivalence (PK) study is conducted using a two-treatment crossover study design in a limited number of volunteers, usually 24 to 36 adults. Alternately, a four-period, replicate design crossover study may also be used. Single doses of the test and reference drug products are administered and blood or plasma levels of the drug are measured over time. Pharmacokinetic parameters characterizing rate and extent of drug absorption are evaluated statistically. The PK parameters of interest are the resulting area under the plasma concentration-

time curve (AUC), calculated to the last measured concentration ($AUC_{(0-t)}$) and extrapolated to infinity ($AUC_{(0-inf)}$), for extent of absorption; and the maximum or peak drug concentrations (Cmax), for rate of absorption. Crossover studies may not be practical in drugs with a long half-life in the body, and a parallel study design may be used instead. Alternate study methods, such as in-vitro studies or equivalence studies with clinical or pharmacodynamic endpoints, are used for drug products where plasma concentrations are not useful to determine delivery of the drug substance to the site of activity (such as inhalers, nasal sprays and topical products applied to the skin).

The statistical methodology for analyzing these bioequivalence studies is called the two one-sided test procedure. Two situations are tested with this statistical methodology. The first of the two one-sided tests determines whether a generic product (test), when substituted for a brand-name product (reference) is significantly less bioavailable. The second of the two one-sided tests determines whether a brand-name product when substituted for a generic product is significantly less bioavailable. Based on the opinions of FDA medical experts, a difference of greater than 20% for each of the above tests was determined to be significant, and therefore, undesirable for all drug products. Numerically, this is expressed as a limit of test-product average/reference-product average of 80% for the first statistical test and a limit of reference-product average/test-product average of 80% for the second statistical test. By convention, all data is expressed as a ratio of the average response (AUC and Cmax) for test/reference, so the limit expressed in the second statistical test is 125% (reciprocal of 80%).

For statistical reasons, all data is log-transformed prior to conducting statistical testing. In practice, these statistical tests are carried out using an analysis of variance procedure (ANOVA) and calculating a 90% confidence interval for each pharmacokinetic parameter (Cmax and AUC). The confidence interval for both pharmacokinetic parameters, AUC and Cmax, must be entirely within the 80% to 125% boundaries cited above. Because the mean of the study data lies in the center of the 90% confidence interval, the mean of the data is usually close to 100% (a test/reference ratio of 1). Different statistical criteria are sometimes used when bioequivalence is demonstrated through comparative clinical trials, pharmacodynamic studies, or comparative in-vitro methodology.

The bioequivalence methodology and criteria described above simultaneously control for both, differences in the average response between test and reference, as well as the precision with which the average response in the population is estimated. This precision depends on the within-subject (normal volunteer or patient) variability in the pharmacokinetic parameters (AUC and Cmax) of the two products and on the number of subjects in the study. The width of the 90% confidence interval is a reflection in part of the within-subject variability of the test and reference products in the bioequivalence study. A test product with no differences in the average response when compared to the reference might still fail to pass the bioequivalence criteria if the variability of one or both products is high and the bioequivalence study has insufficient statistical power (i.e., insufficient number of subjects). Likewise, a test product with low variability may pass the bioequivalence criteria, when there are somewhat larger differences in the average response.

This system of assessing bioequivalence of generic products assures that these substitutable products do not deviate substantially in in-vivo performance from the reference product. The Office of Generic Drugs has conducted two surveys to quantify the differences between generic

and brand name products. The first survey included 224 bioequivalence studies submitted in approved applications during 1985 and 1986. The observed average differences between reference and generic products for AUC was 3.5% (JAMA, Sept. 4, 1987, Vol. 258, No. 9). The second survey included 127 bioequivalence studies submitted to the agency in 273 ANDAs approved in 1997. The three measures reviewed include $AUC_{(0-t)}$, $AUC_{(0-inf)}$, and Cmax. The observed average differences between the reference and generic products were ± 3.47% (SD 2.84) for $AUC_{(0-t)}$, ± 3.25% (SD 2.97) for $AUC_{(0-inf)}$, and ± 4.29% (SD 3.72) for Cmax (JAMA, Dec. 1, 1999, Vol. 282, No. 21).

The primary concern from the regulatory point of view is the protection of the patient against approval of products that are not bioequivalent. The current practice of carrying out two one-sided tests at the 0.05 level of significance ensures that there is no more than a 5% chance that a generic product that is not truly equivalent to the reference will be approved.

Reference Listed Drug (RLD)

A reference listed drug (21 CFR 314.94(a)(3)) means the listed drug identified by FDA as the drug product upon which an applicant relies in seeking approval of its ANDA.

FDA has identified in the Prescription Drug Product and OTC Drug Product Lists those reference listed drugs to which the *in vivo* bioequivalence and, in some instances, the *in vitro* bioequivalence of the applicant's product is compared. By designating a single reference listed drug as the standard to which all generic versions must be shown to be bioequivalent, FDA hopes to avoid possible significant variations among generic drugs and their brand name counterpart. Such variations could result if generic drugs were compared to different reference listed drugs. However, in some instances when listed drugs are approved for a single drug product, a product not designated as the reference listed drug and not shown to be bioequivalent to the reference listed drug may be shielded from generic competition. A firm wishing to market a generic version of a listed drug that is not designated as the reference listed drug may petition the Agency through the Citizen Petition procedure (see 21 CFR 10.25(a) and CFR 10.30). When the Citizen Petition is approved, the second listed drug will be designated as an additional reference listed drug and the petitioner may submit an Abbreviated New Drug Application citing the designated reference listed drug. *Therapeutic Equivalence Evaluations Codes Products meeting necessary bioequivalence requirements* explains the ***AB, AB1, AB2, AB3*** coding system for multisource drug products listed under the same heading with two reference listed drugs.

In addition, there are two situations in which two listed drugs that have been shown to be bioequivalent to each other may both be designated as reference listed drugs. The first situation occurs when the *in vivo* determination of bioequivalence is self-evident and a waiver of *in vivo* determination of bioequivalence may be granted. The second situation occurs when the bioequivalence of two listed drugs may be determined through *in vitro* methodology. The reference listed drug is identified by a "Yes" in the Prescription and Over-the-Counter (OTC) Drug Product Lists and is identified in the printed version by a "+". These identified reference listed drugs represent the best judgment of the Division of Bioequivalence at this time. The Prescription and OTC Drug Product Lists identify reference drugs for oral dosage forms, injectables, ophthalmics, otics, and topical products. It is recommended that a firm planning to conduct an *in vivo* bioequivalence study, or planning to manufacture a batch of a drug product for which an *in vivo* waiver of bioequivalence will be requested, contact the Division of

Bioequivalence, Office of Generic Drugs, to confirm the appropriate reference listed drug.

Acyclovir 200mg Tablet-Reference Listed Drug. Novopharm's single source acyclovir tablets have been declared to be a reference listed drug for the 200 mg tablet in addition to the acylcovir (Zovirax) 800 mg tablet of the innovator. A generic firm wishing to submit an ANDA for a duplicate of the 200 mg acyclovir tablet will be eligible for a waiver of the *in vivo* determination of bioequivalence (1) if their product is proportionally similar in its active and inactive ingredients to their own 800 mg acyclovir tablet and (2) by doing an acceptable comparative dissolution test (dissolution profile) against Novopharm's 200 mg acyclovir reference listed drug.

Before a waiver of the *in vivo* determination of bioequivalence can be granted for the 200 mg acyclovir tablet, the generic firm must have completed an acceptable fasting and fed study comparing their acyclovir 800 mg tablet against the Zovirax 800 mg tablet.

For further information on the study designs, you should contact the Division of Bioequivalence, Office of Generic Drugs.

General Policies and Legal Status

The List contains public information and advice. It does not mandate the drug products which may be purchased, prescribed, dispensed, or substituted for one another, nor does it, conversely, mandate the products that should be avoided. To the extent that the List sets forth FDA's evaluations of the therapeutic equivalence of drug products that have been approved, it contains FDA's advice to the public, to practitioners and to the states regarding drug product selection. These evaluations do not constitute determinations that any product is in violation of the Act or that any product is preferable to any other. Therapeutic equivalence evaluations are a scientific judgment based upon evidence, while generic substitution may involve social and economic policy administered by the states, intended to reduce the cost of drugs to consumers. To the extent that the List identifies drug products approved under Section 505 of the Act, it sets forth information that the Agency is required to publish and that the public is entitled to under the Freedom of Information Act. Exclusion of a drug product from the List does not necessarily mean that the drug product is either in violation of Section 505 of the Act, or that such a product is not safe or effective, or that such a product is not therapeutically equivalent to other drug products. Rather, the exclusion is based on the fact that FDA has not evaluated the safety, effectiveness, and quality of the drug product.

Practitioner/User Responsibilities

Professional care and judgment should be exercised in using the List. Evaluations of therapeutic equivalence for prescription drugs are based on scientific and medical evaluations by FDA. Products evaluated as therapeutically equivalent can be expected, in the judgment of FDA, to have equivalent clinical effect and no difference in their potential for adverse effects when used under the conditions of their labeling. However, these products may differ in other characteristics such as shape, scoring configuration, release mechanisms, packaging, excipients (including colors, flavors, preservatives), expiration date/time, and, in some instances, labeling. If products with such differences are substituted for each other, there is a potential for patient confusion due to differences in color or shape of tablets, inability to provide a given dose using a partial tablet if the proper scoring configuration is not available, or decreased patient acceptance

of certain products because of flavor. There may also be better stability of one product over another under adverse storage conditions, or allergic reactions in rare cases due to a coloring or a preservative ingredient, as well as differences in cost to the patient.

FDA evaluation of therapeutic equivalence in no way relieves practitioners of their professional responsibilities in prescribing and dispensing such products with due care and with appropriate information to individual patients. In those circumstances where the characteristics of a specific product, other than its active ingredient, are important in the therapy of a particular patient, the physician's specification of that product is appropriate. Pharmacists must also be familiar with the expiration dates/times and labeling directions for storage of the different products, particularly for reconstituted products, to assure that patients are properly advised when one product is substituted for another.

Multisource and single-source drug products. FDA has evaluated for therapeutic equivalence only multisource prescription drug products, which in most instances means those pharmaceutical equivalents available from more than one manufacturer. A therapeutic equivalence code is included for such products. Those products with approved applications that are single-source (i.e., there is only one approved product available for that active ingredient, dosage form and route of administration) are also included on the List, but no therapeutic equivalence code is included with such products. Any drug product in the List repackaged and/or distributed by other than the application holder is considered to be therapeutically equivalent to the application holder's drug product even if the application holder's drug product is single source or coded as non-equivalent (e.g., **BN**). Also, although not identified in the List, distributors or repackagers of an application holder's drug product are considered to have the same code as the application holder. The details of these codes and the policies underlying them are discussed in *Therapeutic Equivalence Evaluations Codes*.

Products on the List are identified by the names of the holders of approved applications (applicants) who may not necessarily be the manufacturer of the product. The applicant may have had its product manufactured by a contract manufacturer and may simply be distributing the product for which it has obtained approval. In most instances, however, the manufacturer of the product is also the applicant. The name of the manufacturer is permitted by regulation to appear on the label, even when the manufacturer is not the marketer.

Although the products on the List are identified by the names of the applicants, circumstances, such as changing corporate ownership, have sometimes made identification of the applicant difficult. The Agency believes, based on continuing document review and communication with firms, that the applicant designations on the List are, in most cases, correct.

To relate firm name information on a product label to that on the List, the following should be noted: the applicant's name always appears on the List. This applies whether the applicant (firm name on the Form FDA 356h in the application) is the marketer (firm name in largest letters on the label) or not. However, the applicant's name may not always appear on the label of the product.

If the applicant is the marketer, its name appears on the List and on the label; if the applicant is not the marketer, and the Agency is aware of a corporate relationship (e.g., parent and subsidiary) between the applicant and the marketer, the name of the applicant appears on the List and both firm names may appear on the label. If there is no known corporate relationship between the

applicant and the marketer, the applicant's name appears on the List; however, unless the applicant is the manufacturer, packager, or distributor, the applicant's name may not appear on the label. In this case, the practitioner, from labeling alone, will not be able to relate the marketed product to an applicant cited in the List, and hence to a specific approved drug product. In such cases, to assure that the product in question is the subject of an approved application, the firm named on the label should be contacted.

To relate trade name (proprietary name) information on a product label to that on the List, the following should be noted: if the applicant is the marketer, its name appears on the List and on the label; if the Agency is aware of a corporate relationship between the applicant and the marketer, the trade name (proprietary name) of the drug product (established drug name if no trade name exists) appears on the List. If a corporate relationship exists between an application holder and a marketer and both firms are distributing the drug product, the FDA reserves the right to select the trade name of either the marketer or the application holder to appear on the List. If there is no known corporate relationship between the applicant and the marketer, the established drug name appears on the List.

Every product on the List is subject at all times to regulatory action. From time to time, approved products may be found in violation of one or more provisions of the Act. In such circumstances, the Agency will commence appropriate enforcement action to correct the violation, if necessary, by securing removal of the product from the market by voluntary recall, seizure, or other enforcement actions. Such regulatory actions are, however, independent of the inclusion of a product on the List. The main criterion for inclusion of a product is that it has an application with an effective approval that has not been withdrawn for safety or efficacy reasons. FDA believes that retention of a violative product on the List will not have any significant adverse health consequences, because other legal mechanisms are available to the Agency to prevent the product's actual marketing. FDA may however, change a product's therapeutic equivalence rating if the circumstances giving rise to the violation change or otherwise call into question the data upon which the Agency's assessment of whether a product meets the criteria for therapeutic equivalence was made.

Therapeutic Equivalence Evaluations Codes

The coding system for therapeutic equivalence evaluations is constructed to allow users to determine quickly whether the Agency has evaluated a particular approved product as therapeutically equivalent to other pharmaceutically equivalent products (first letter) and to provide additional information on the basis of FDA's evaluations (second letter). With few exceptions, the therapeutic equivalence evaluation date is the same as the approval date.

The two basic categories into which multisource drugs have been placed are indicated by the first letter as follows:

A Drug products that FDA considers to be therapeutically equivalent to other pharmaceutically equivalent products, i.e., drug products for which:

(1) there are no known or suspected bioequivalence problems. These are designated **AA**, **AN**, **AO**, **AP**, or **AT**, depending on the dosage form; or

(2) actual or potential bioequivalence problems have been resolved with adequate *in vivo* and/or *in vitro* evidence supporting bioequivalence. These are designated **AB**.

B Drug products that FDA at this time, considers NOT to be therapeutically equivalent to other pharmaceutically equivalent products, i.e.,

drug products for which actual or potential bioequivalence problems have not been resolved by adequate evidence of bioequivalence. Often the problem is with specific dosage forms rather than with the active ingredients. These are designated **BC, BD, BE, BN, BP, BR, BS, BT, BX,** or **B***.

Individual drug products have been evaluated as therapeutically equivalent to the reference product in accordance with the definitions and policies outlined below:

■ "A" CODES

Drug products that are considered to be therapeutically equivalent to other pharmaceutically equivalent products.

"**A**" products are those for which actual or potential bioequivalence problems have been resolved with adequate *in vivo* and/or *in vitro* evidence supporting bioequivalence. Drug products designated with an "**A**" code fall under one of two main policies:

(1) for those active ingredients or dosage forms for which no *in vivo* bioequivalence issue is known or suspected, the information necessary to show bioequivalence between pharmaceutically equivalent products is presumed and considered self-evident based on other data in the application for some dosage forms (e.g., solutions) or satisfied for solid oral dosage forms by a showing that an acceptable *in vitro* dissolution standard is met. A therapeutically equivalent rating is assigned such products so long as they are manufactured in accordance with Current Good Manufacturing Practice regulations and meet the other requirements of their approved applications (these are designated **AA, AN, AO, AP,** or **AT**, depending on the dosage form, as described below); or

(2) for those DESI drug products containing active ingredients or dosage forms that have been identified by FDA as having actual or potential bioequivalence problems, and for post-1962 drug products in a dosage form presenting a potential bioequivalence problem, an evaluation of

therapeutic equivalence is assigned to pharmaceutical equivalents only if the approved application contains adequate scientific evidence establishing through *in vivo* and/or *in vitro* studies the bioequivalence of the product to a selected reference product (these products are designated as **AB**).

There are some general principles that may affect the substitution of pharmaceutically equivalent products in specific cases. Prescribers and dispensers of drugs should be alert to these principles so as to deal appropriately with situations that require professional judgment and discretion.

There may be labeling differences among pharmaceutically equivalent products that require attention on the part of the health professional. For example, pharmaceutically equivalent powders to be reconstituted for administration as oral or injectable liquids may vary with respect to their expiration time or storage conditions after reconstitution. An FDA evaluation that such products are therapeutically equivalent is applicable only when each product is reconstituted, stored, and used under the conditions specified in the labeling of that product.

The Agency will use notes in this publication to point out special situations such as potential differences between two drug products that have been evaluated as bioequivalent and otherwise therapeutically equivalent, when they should be brought to the attention of health professionals. These notes are contained in *Description of Special Situations*.

For example, in rare instances, there may be variations among therapeutically equivalent products in their use or in conditions of administration. Such differences may be due to patent or exclusivity rights associated with such use. When such variations may, in the Agency's opinion, affect prescribing or substitution decisions by health professionals, a note will be added to *Description of Special Situations*.

Also, occasionally a situation may arise in which changes in a listed drug product after its approval (for example, a change in dosing interval) may have an impact on the substitutability of already approved generic versions of that product that were rated by the Agency as therapeutically equivalent to the listed product. When such changes in the listed drug product are considered by the Agency to have a significant impact on therapeutic equivalence, the Agency will change the therapeutic equivalence ratings for other versions of the drug product unless the manufacturers of those other versions of the product provide additional information to assure equivalence under the changed conditions. Pending receipt of the additional data, the Agency may add a note to *Description of Special Situations*, or, in rare cases, may even change the therapeutic equivalence rating.

In some cases (e.g., Isolyte® S w/ Dextrose 5% in Plastic Container and Plasma-Lyte® 148 and Dextrose 5% in Plastic Container), closely related products are listed as containing the same active ingredients, but in somewhat different amounts. In determining which of these products are pharmaceutically equivalent, the Agency has considered products to be pharmaceutically equivalent with labeled strengths of an ingredient that do not vary by more than 1%.

Different salts and esters of the same therapeutic moiety are regarded as pharmaceutical alternatives. For the purpose of this publication, such products are not considered to be

therapeutically equivalent. There are no instances in this List where pharmaceutical alternatives are evaluated or coded with regard to therapeutic equivalence. Anhydrous and hydrated entities, as well as different polymorphs, are considered pharmaceutical equivalents and must meet the same standards and, where necessary, as in the case of ampicillin/ampicillin trihydrate, their equivalence is supported by appropriate bioavailability/bioequivalence studies.

The codes in this book are not intended to preclude health care professionals from converting pharmaceutically different concentrations into pharmaceutical equivalents using accepted professional practice.

Where package size variations have therapeutic implications, products so packaged have not been considered pharmaceutically equivalent. For example, some oral contraceptives are supplied in 21-tablet and 28-tablet packets; the 28-tablet packets contain 7 placebo or iron tablets. These two packaging configurations are not regarded as pharmaceutically equivalent; thus, they are not designated as therapeutically equivalent.

Preservatives may differ among some therapeutically equivalent drug products. Differences in preservatives and other inactive ingredients do not affect FDA's evaluation of therapeutic equivalence except in cases where these components may influence bioequivalence or routes of administration.

The specific sub-codes for those drugs evaluated as therapeutically equivalent and the policies underlying these sub-codes follow:

AA Products in conventional dosage forms not presenting bioequivalence problems

Products coded as **AA** contain active ingredients and dosage forms that are not regarded as presenting either actual or potential bioequivalence problems or drug quality or standards issues. However, all oral dosage forms must, nonetheless, meet an appropriate *in vitro* bioequivalence standard that is acceptable to the Agency in order to be approved.

AB, AB1, AB2, AB3... Products meeting necessary bioequivalence requirements

Multisource drug products listed under the same heading (i.e., identical active ingredients(s), dosage form, and route(s) of administration) and having the same strength (see *Therapeutic Equivalence-Related Terms, Pharmaceutical Equivalents)* generally will be coded **AB** if a study is submitted demonstrating bioequivalence.

In certain instances, a number is added to the end of the AB code to make a three character code (i.e., AB1, AB2, AB3, etc.). Three-character codes are assigned only in situations when more than one reference listed drug of the same strength has been designated under the same heading. Two or more reference listed drugs are generally selected only when there are at least two potential reference drug products which are not bioequivalent to each other. If a study is submitted that demonstrates bioequivalence to a specific listed drug product, the generic product will be given the same three-character code as the

reference listed drug it was compared against. For example, Adalat® CC (Miles) and Procardia XL® (Pfizer), extended-release tablets, are listed under the active ingredient nifedipine. These drug products, listed under the same heading, are not bioequivalent to each other. Generic drug products deemed by FDA to be bioequivalent to Adalat® CC and Procardia XL® have been approved, Adalat® CC and Procardia XL® have been assigned ratings of AB1 and AB2, respectively. The generic drug products bioequivalent to Adalat® CC would be assigned a rating of AB1 and those bioequivalent to Procardia XL® would be assigned a rating of AB2. (The assignment of an AB1 or AB2 rating to a specific product does not imply product preference.) Even though drug products of distributors and/or repackagers are not included in the List, they are considered therapeutically equivalent to the application holder's drug product if the application holder's drug product is rated either with an AB or three-character code or is single source in the List. Drugs coded as AB under a heading are considered therapeutically equivalent only to other drugs coded as AB under that heading. Drugs coded with a three-character code under a heading are considered therapeutically equivalent only to other drugs coded with the same three-character code under that heading.

AN Solutions and powders for aerosolization

Uncertainty regarding the therapeutic equivalence of aerosolized products arises primarily because of differences in the drug delivery system. Solutions and powders intended for aerosolization that are marketed for use in any of several delivery systems are considered to be pharmaceutically and therapeutically equivalent and are coded **AN**. Those products that are compatible only with a specific delivery system or those products that are packaged in and with a specific delivery system are coded **BN**, unless they have met an appropriate bioequivalence standard. Solutions or suspensions in a specific delivery system will be coded **AN** if the bioequivalence standard is based upon *in vitro* methodology, if bioequivalence needs to be demonstrated by *in vivo* methodology then the drug products will be coded **AB**.

AO Injectable oil solutions

The absorption of drugs in injectable (parenteral) oil solutions may vary substantially with the type of oil employed as a vehicle and the concentration of the active ingredient. Injectable oil solutions are therefore considered to be pharmaceutically and therapeutically equivalent only when the active ingredient, its concentration, and the type of oil used as a vehicle are all identical.

AP Injectable aqueous solutions and, in certain instances, intravenous non-aqueous solutions

It should be noted that even though injectable (parenteral) products under a specific listing may be evaluated as therapeutically equivalent, there may be important differences among the products in the general category, *Injectable; Injection*. For example, some injectable products that are rated therapeutically equivalent are labeled for different routes of administration. In addition, some products evaluated as therapeutically equivalent may have different preservatives or no preservatives at all. Injectable products available as dry powders for reconstitution, concentrated sterile solutions for dilution, or sterile solutions ready for injection are all considered to be pharmaceutically and therapeutically equivalent provided they are designed to produce the same concentration prior to injection and are similarly labeled. Consistent with accepted professional practice, it is the responsibility of the prescriber, dispenser, or individual administering the product to be familiar with a product's labeling to assure that it is given only by the route(s) of administration stated in the labeling.

Certain commonly used large volume intravenous products in glass containers are not included on the List (e.g., dextrose injection 5%, dextrose injection 10%, sodium chloride injection 0.9%) since these products are on the market without FDA approval and the FDA has not published conditions for marketing such parenteral products under approved NDAs. When packaged in plastic containers, however, FDA regulations require approved applications prior to marketing. Approval then depends on, among other things, the extent of the available safety data involving the specific plastic component of the product. All large volume parenteral products are manufactured under similar standards, regardless of whether they are packaged in glass or plastic. Thus, FDA has no reason to believe that the packaging container of large volume parenteral drug products that are pharmaceutically equivalent would have any effect on their therapeutic equivalence.

The strength of parenteral drugs products is defined as the total drug content of the container. Until recently the strength of liquid parenteral drug products in the Orange Book have not been displayed. The concentration of the liquid parenteral drug product in the Orange Book has been shown as xmg/ml. The amount of dry powder or freeze dried powder in a container has always been identified as the strength.

With the finalization of the Waxman-Hatch amendments that characterized each strength of a drug product as a listed drug it became evident that the format of the Orange Book should be changed to reflect each strength of a parenteral solution. To this end the OGD has started to display the strength of all new approvals of parenteral solutions. Previously we would have displayed only the concentration of an approved parenteral solution, e.g. 50mg/ml. If this drug product had a 20 ml and 60 ml container approved the two products would be shown as 1Gm / 20ml (50mg/ml) and 3Gm / 60ml (50mg/ml).

AT Topical products

There are a variety of topical dosage forms available for dermatologic, ophthalmic, otic, rectal, and vaginal administration, including solutions, creams, ointments, gels, lotions, pastes, sprays, and suppositories. Even though different topical dosage forms may contain the same active ingredient and potency, these dosage forms are not

considered pharmaceutically equivalent. Therefore, they are not considered therapeutically equivalent. All solutions and DESI drug products containing the same active ingredient in the same topical dosage form for which a waiver of *in vivo* bioequivalence has been granted and for which chemistry and manufacturing processes are adequate to demonstrate bioequivalence, are considered therapeutically equivalent and coded **AT**. Pharmaceutically equivalent topical products that raise questions of bioequivalence, including all post-1962 non-solution topical drug products, are coded **AB** when supported by adequate bioequivalence data, and **BT** in the absence of such data.

"B" CODES

Drug products that FDA, at this time, considers not to be therapeutically equivalent to other pharmaceutically equivalent products.

"**B**" products, for which actual or potential bioequivalence problems have not been resolved by adequate evidence of bioequivalence, often have a problem with specific dosage forms rather than with the active ingredients. Drug products designated with a "**B**" code fall under one of three main policies:

(1) the drug products contain active ingredients or are manufactured in dosage forms that have been identified by the Agency as having documented bioequivalence problems or a significant potential for such problems and for which no adequate studies demonstrating bioequivalence have been submitted to FDA; or

(2) the quality standards are inadequate or FDA has an insufficient basis to determine therapeutic equivalence; or

(3) the drug products are under regulatory review.

The specific coding definitions and policies for the "**B**" sub-codes are as follows:

B* Drug products requiring further FDA investigation and review to determine therapeutic equivalence

The code **B*** is assigned to products previously assigned an **A** or **B** code when FDA receives new information that raises a significant question regarding therapeutic equivalence that can be resolved only through further Agency investigation and/or review of data and information submitted by the applicant. The **B*** code signifies that the Agency will take no position regarding the therapeutic equivalence of the product until the Agency completes its investigation and review.

BC Extended-release dosage forms (capsules, injectables and tablets)

An extended-release dosage form is defined by the official compendia as one that allows at least a twofold reduction in dosing frequency as compared to that drug presented as a conventional dosage form (e.g., as a solution or a prompt drug-releasing, conventional solid dosage form).

Although bioavailability studies have been conducted on these dosage forms, they may be subject to bioavailability differences, primarily because firms developing extended-release products for the same active ingredient rarely employ the same formulation approach. FDA, therefore, does not consider different extended-release dosage forms containing the same active ingredient in equal strength to be therapeutically equivalent unless equivalence between individual products in both rate and extent has been specifically demonstrated through appropriate bioequivalence studies. Extended-release products for which such bioequivalence data have not been submitted are coded **BC**, while those for which such data are available have been coded **AB**.

BD Active ingredients and dosage forms with documented bioequivalence problems

The **BD** code denotes products containing active ingredients with known bioequivalence problems and for which adequate studies have not been submitted to FDA demonstrating bioequivalence. Where studies showing bioequivalence have been submitted, the product has been coded **AB**.

BE Delayed-release oral dosage forms

A delayed-release dosage form is defined by the official compendia as one that releases a drug (or drugs) at a time other than promptly after administration. Enteric-coated articles are delayed-release dosage forms.

Drug products in delayed-release dosage forms containing the same active ingredients are subject to significant differences in absorption. Unless otherwise specifically noted, the Agency considers different delayed-release products

containing the same active ingredients as presenting a potential bioequivalence problem and codes these products **BE** in the absence of *in vivo* studies showing bioequivalence. If adequate *in vivo* studies have demonstrated the bioequivalence of specific delayed-release products, such products are coded **AB**.

BN Products in aerosol-nebulizer drug delivery systems

This code applies to drug solutions or powders that are marketed only as a component of, or as compatible with, a specific drug delivery system. There may, for example, be significant differences in the dose of drug and particle size delivered by different products of this type. Therefore, the Agency does not consider different metered aerosol dosage forms containing the same active ingredient(s) in equal strengths to be therapeutically equivalent unless the drug products meet an appropriate bioequivalence standard.

BP Active ingredients and dosage forms with potential bioequivalence problems

FDA's bioequivalence regulations (21 CFR 320.33) contain criteria and procedures for determining whether a specific active ingredient in a specific dosage form has a potential for causing a bioequivalence problem. It is FDA's policy to consider an ingredient meeting these criteria as having a potential bioequivalence problem even in the absence of positive data demonstrating inequivalence. Pharmaceutically equivalent products containing these ingredients in oral dosage forms are coded **BP** until adequate *in vivo* bioequivalence data are submitted. Injectable suspensions containing an active ingredient suspended in an aqueous or oleaginous vehicle have also been coded **BP**. Injectable suspensions are subject to bioequivalence problems because differences in particle size, polymorphic structure of the suspended active ingredient, or the suspension formulation can significantly affect the rate of release and absorption. FDA does not consider pharmaceutical equivalents of these products bioequivalent without adequate evidence of bioequivalence, such products would be coded AB.

BR Suppositories or enemas that deliver drugs for systemic absorption

The absorption of active ingredients from suppositories or enemas that are intended to have a systemic effect (as distinct from suppositories administered for local effect) can vary significantly from product to product. Therefore, FDA considers pharmaceutically equivalent systemic suppositories or enemas bioequivalent only if *in vivo* evidence of bioequivalence is available. In those cases where *in vivo* evidence is available, the product is coded **AB**. If such evidence is not available, the products are coded **BR**.

BS Products having drug standard deficiencies

If the drug standards for an active ingredient in a particular dosage form are found by FDA to be deficient so as to prevent an FDA evaluation of either pharmaceutical or therapeutic equivalence, all drug products containing that active ingredient in that dosage form are coded **BS**. For example, if the standards permit a wide variation in pharmacologically active components of the active ingredient such that pharmaceutical equivalence is in question, all products containing that active ingredient in that dosage form are coded **BS**.

BT Topical products with bioequivalence issues

This code applies mainly to post-1962 dermatologic, ophthalmic, otic, rectal, and vaginal products for topical administration, including creams, ointments, gels, lotions, pastes, and sprays, as well as suppositories not intended for systemic drug absorption. Topical products evaluated as having acceptable clinical performance, but that are not bioequivalent to other pharmaceutically equivalent products or that lack sufficient evidence of bioequivalence, will be coded **BT**.

BX Drug products for which the data are insufficient to determine therapeutic equivalence

The code **BX** is assigned to specific drug products for which the data that have been reviewed by the Agency are insufficient to determine therapeutic equivalence under the policies stated in this document. In these situations, the drug products are presumed to be therapeutically inequivalent until the Agency has determined that there is adequate information to make a full evaluation of therapeutic equivalence.

Description of Special Situations

Certain drugs present special situations that deserve a more complete explanation than can be provided by the two-letter codes used in the List. These drugs have particular problems with standards of identity, analytical methodology, or bioequivalence that are in the process of resolution. The following drugs are in this category:

Amino Acid and Protein Hydrolysate Injections. These products differ in the amount and kinds of amino acids they contain and, therefore, are not considered pharmaceutical equivalents. For this reason, these products are not considered therapeutically equivalent. At the same time, the Agency believes that it is appropriate to point out that where nitrogen balance is the sole therapeutic objective and individual amino acid content is not a consideration, pharmaceutical

alternatives with the same total amount of nitrogen content may be considered therapeutically equivalent.

Ribavirin 200mg Oral Capsule Indicated for use and comarketed with interferon alfa-2b, recombinant (Intron A), as Rebetron Combination Therapy.

Follitropin Alfa and Beta Based on available data derived from physico-chemical tests and bioassay, follitropin alfa and follitropin beta are indistinguishable.

Gaviscon®. Gaviscon® is an OTC product which has been marketed since September 1970. The active ingredients in this product, aluminum hydroxide and magnesium trisilicate, were reviewed by the Agency's OTC Antacid Panel and were considered to be safe and effective ingredients (Category I) by that Panel. However, the tablet failed to pass the antacid test which is required of all antacid products. The Agency, therefore, placed the tablet in Category III for lack of effectiveness. A full NDA with clinical studies was submitted by Marion Laboratories, Inc., and approved by FDA on December 9, 1983. Gaviscon® 's activity in treating reflux acidity is made possible by the physical-chemical properties of the inactive ingredients, sodium bicarbonate and alginic acid. Therefore, *all ANDAs which cite Gaviscon® tablets as the listed drug must contain the inactive ingredients sodium bicarbonate and alginic acid.* A full NDA will be required to support the effectiveness of the drug product if different inactive ingredients are to be substituted for sodium bicarbonate or alginic acid or if different proportions of these ingredients are to be used.

Patent Certification(s) Reference Listed Drug based upon a suitability petition. An abbreviated new drug application that refers to a Reference Listed Drug (RLD) approved pursuant to a suitability petition must demonstrate that the proposed product is bioequivalent to the RLD, and it must include appropriate patent certification(s) and an exclusivity statement with respect to the listed drug which served as the basis for the approved suitability petition.

Waived exclusivity. If a new drug application (NDA) submitted under section 505(b) of the Federal Food, Drug, and Cosmetic Act (Act) qualifies for exclusivity under sections 505(c)(3)(D) and 505(j)(5)(D), the exclusivity is listed in the Patent and Exclusivity Section of the Orange Book. If a drug product has received this exclusivity, the FDA will delay the approval of a 505(b)(2) application or an abbreviated new drug application (ANDA) under section 505(j) of the Act until the expiration of the exclusivity. If the listed drug is also protected by one or more patents, the approval date for the 505(b)(2) application or ANDA will be determined by the latest expiring patent or exclusivity listed in the Orange Book.

However, the holder of the NDA may waiver its exclusivity as to any or all 505(b)(2) and ANDA applications referencing the protected drug product. If an NDA sponsor waivers its right to the exclusivity protection, qualified 505(b)(2) or ANDA applications may be approved without regard to the NDA holder's exclusivity. An NDA for which the holder has waived its exclusivity as to all 505(b)(2) and ANDA applications will be coded with a W in the Patent and Exclusivity Section of the Orange Book and be referred to this section. The applicant referencing this listed drug should indicate in the exclusivity statement that the holder of the listed drug has waived its exclusivity.

Therapeutic Equivalence Code Change for a Drug Entity

The Agency will use the following procedures when, in response to a petition or on its own initiative, it is considering a change in the therapeutic equivalence code for approved multi-source drug products. Such changes will generally occur when the Agency becomes aware of new scientific information affecting the therapeutic equivalence of an entire category of drug products in the List (e.g., information concerning the active ingredient or the dosage form), rather than information concerning a single drug product within the category. These procedures will be used when a change in therapeutic equivalence code is under consideration for all drug products found in the Prescription Drug Product List under a specific drug entity and dosage form. The change may be from the code signifying that the drug does not present a bioequivalence problem (e.g., **AA**) to a code signifying a bioequivalence problem (e.g., **BP**), or vice versa. This procedure does not apply to a change of a particular product code (e.g., a change from **BP** to **AB** or from **AB** to **BX**).

Before making a change in a therapeutic equivalence code for an entire category of drugs, the Agency will announce in the *Introduction* that it is considering the change, and will invite comment. Comments, along with scientific data, may be sent to the Director, Division of Bioequivalence, Office of Generic Drugs, Center for Drug Evaluation and Research, (MPN-2) HFD-650, 7500 Standish Place, Rockville, MD 20855. The comment period will generally be 60 days in length, and the closing date for comments will be listed in the description of the proposed change for each drug entity.

The most useful type of scientific data submission is an *in vivo* bioavailability/bioequivalence study conducted on batches of the subject drug products. These submissions should present a full description of the analytical procedures and equipment used, a validation of the analytical methodology, including the standard curve, a description of the method of calculating results, and a description of the pharmacokinetic and statistical models used in analyzing the data. Anecdotal or testimonial information is the least useful to the Agency, and such submissions are discouraged. Copies of supporting reports published in the scientific literature or unpublished material, however, are welcome.

Change of the Therapeutic Equivalence Evaluation for a Single Product

The aforementioned procedure does not apply to a change in a single drug product code. For example, a change in a single drug product's code from **BP** to **AB** as a result of the submission of a bioequivalence study ordinarily will not be the subject of notice and comment. Likewise, a change in a single drug product's code from **AB** to **BX** (e.g., as a result of new information raising a significant question as to bioequivalence) does not require notice and comment. The Agency's responsibility to provide the public with the Agency's most current information related to therapeutic equivalence may require a change in a drug product's code prior to any formal notice and opportunity for the applicant to be heard. The publication in the *Federal Register* of a proposal to withdraw approval of a drug product will ordinarily result in a change in a product's code from **AB** to **BX** if this action has not already been taken.

Availability of Internal Policy and Procedure Guides

The Office of Generic Drugs maintains internal policy and procedure guides. Although these guides are designed for Office personnel and are subject to change without public notice, they are available to members of the public who may wish to know more about the Office's policies and procedures. Copies of these guides may be obtained from the FDA, Center for Drug Evaluation and Research, HFD-240, Office of Training and Communications, Division of Drug Information, 5600 Fishers Lane, Rockville, MD 20857. The Agency welcomes public comment on the policies, procedures, and practices employed in the approval of generic drugs. Such comments may be sent to the Director, Office of Generic Drugs, (MPN-2) HFD-600, 7500 Standish Place, Rockville, MD 20855.

Discontinued Section

Those drug products in the Discontinued Section of the Orange Book in which a determination has already been made that the products were not marketed or withdrawn for safety or efficacy reasons have been designated by the symbol "*". Those drug products with the symbol "*" are only reflective of citizen petitions approved since 1995.

The identification of these drug products in the Discontinued Section of the Orange Book with the symbol "*" should avoid the submission of multiple citizen petitions for the same drug product.

HOW TO USE THE DRUG PRODUCT LISTS

Key Sections for Using the Drug Product Lists

This publication contains Drug Product Lists and lists of abbreviations and terms which facilitate their use.

Drug Product Lists. The Prescription Drug Product, OTC Drug Product and Discontinued Drug Product sections may be searched by one of four criteria: (1) Active Ingredient; (2) Applicant Holder; (3) Proprietary Name; and (4) Application Number. Regardless of which criterion is searched, the results are sorted first alphabetically by the active ingredient, then by dosage form, and then by the applicant holder for single and multiple ingredient drug products. The fields displayed may vary depending on the query that is chosen. In addition, the queries may contain the following fields: therapeutic equivalence code; reference listed drug (RLD) designation; and strength(s). Note, the therapeutic equivalence code field will appear in the Prescription Drug Product List only. The reference listed drug field will only appear in Prescription Drug Product and OTC Drug Product Lists. The Discontinued Drug Product List does not contain either a therapeutic equivalence code or a reference listed drug field. In addition, the query results will provide the user with an option to link to each NDA to obtain drug product-specific information. The following fields will be provided for each NDA: the application number and a three-digit drug product number (FDA internal computer data use only); approval dates for those drug products approved on or after January 1, 1982; Rx/OTC/DISCN designation; and patent and exclusivity information.

If a prescription drug product is available from more than one source (multisource), a therapeutic equivalence code will be displayed. If a product is therapeutically equivalent to one or more products or to an appropriate reference, it will be designated with a code beginning with "**A**".

Active ingredient headings for multiple ingredient (combination) drug products are arranged alphabetically. For purposes of this publication, this alphabetical sort takes precedence over United States Pharmacopeia official monograph order (i.e., Reserpine, Hydralazine Hydrochloride, Hydrochlorothiazide). For example, product information labeled as Reserpine, Hydrochlorothiazide and Hydralazine Hydrochloride appears under the active ingredient heading *Hydralazine Hydrochloride; Hydrochlorothiazide; Reserpine.* For combination drug products, the ingredient strengths are separated by semicolons. Available strengths of the dosage form from an applicant appear as separate entries.

Therapeutic equivalence or inequivalence for prescription products is determined on the basis of the therapeutic equivalence codes provided within that specific dosage form. The Discontinued Drug Product List contains approved products that have never been marketed, have been discontinued from marketing, or have had their approvals withdrawn for other than safety or efficacy reasons subsequent to being discontinued from marketing.

Uniform Terms. To improve readability, uniform terms are used to designate dosage forms, routes of administration, and abbreviations used to express strengths. These terms are listed in the Appendix. In some cases, the terms used may differ from those used in product labels and other labeling.

PATENT AND EXCLUSIVITY INFORMATION ADDENDUM

This *Addendum* identifies drugs that qualify under the Drug Price Competition and Patent Term Restoration Act (1984 Amendments) for periods of exclusivity, during which abbreviated new drug applications (ANDAs) and applications described in Section 505(b)(2) of the Federal Food, Drug, and Cosmetic Act (the Act) for those drug products may, in some instances, not be submitted or made effective as described below, and provides patent information concerning the listed drug products. Those drugs that have qualified for Orphan Drug Exclusivity pursuant to Section 527 of the Act and those drugs that have qualified for Pediatric Exclusivity pursuant to Section 505A are also included in this *Addendum*. For an explanation of the codes used in the *Addendum*, see the *Patent and Exclusivity Terms* page. Exclusivity prevents the submission or effective approval of ANDAs or applications described in Section 505(b)(2) of the Act. It does not prevent the submission or approval of a second full NDA. Applications qualifying for periods of exclusivity are:

(1) A new drug application approved after September 24, 1984, for a

drug product all active ingredients (including any ester or salt of the active ingredient) of which had never been approved in any other new drug application under Section 505(b) of the Act. No subsequent ANDA or application described in Section 505(b)(2) of the Act for the same drug may be *submitted* for a period of *five years* from the date of approval of the original application, except that such an application may be *submitted* after *four years* if it contains a certification that a patent claiming the drug is invalid or will not be infringed by the product for which approval is sought.

(2) A new drug application approved after September 24, 1984, for a drug product containing an active ingredient (including any ester or salt of that active ingredient) that has been approved in an earlier new drug application and that includes reports of new clinical investigations (other than bioavailability studies). Such investigations must have been conducted or sponsored by the applicant and must have been essential to approval of the application. If these requirements are met, the approval of a subsequent ANDA or an application described in Section 505(b)(2) of the Act may not be *made effective* for the same drug or use, if for a new indication, before the expiration of *three years* from the date of approval of the original application. If an applicant has exclusivity for a new use or indication, this does not preclude the approval of an ANDA application or 505(b)(2) application for the drug product with indications not covered by the exclusivity.

(3) A supplement to a new drug application for a drug containing a previously approved active ingredient (including any ester or salt of the active ingredient) approved after September 24, 1984, that contains reports of new clinical investigations (other than bioavailability studies) essential to the approval of the supplement and conducted or sponsored by the applicant. The approval of a subsequent application for a change approved in the supplement may not be *made effective* for *three years* from the date of approval of the original supplement.

The Act requires that patent information must now be filed with all newly submitted Section 505 drug applications, and that no NDA may be approved after September 24, 1984, without the submission of pertinent patent information to the Agency. The patent numbers and the expiration dates of appropriate patents claiming drug products that are the subject of approved applications will be published in this *Addendum.* Patent information on unapproved applications or on patents beyond the scope of the Act (i.e., process or manufacturing patents) will not be published.

The patents that FDA regards as covered by the statutory provisions for submission of patent information are: patents that claim the active ingredient or ingredients; drug product patents, which include formulation/composition patents; and use patents for a particular approved indication or method of using the product. NDA holders or applicants amending or supplementing applications with formulation/ composition patent information are asked to declare that the patent(s) is appropriate for publication and refers to an approved product or one for which approval is being sought. The Agency asks all applicants or application holders with use patents to provide information as to the approved indications or uses covered by such patents.

This information will be included in the List as it becomes available.

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Date created: February 26, 2004

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ATTACHMENT G

Evidence for the primary role of anagrelide's major metabolite, 3-hydroxy anagrelide in the drug's clinical activity.

This represents a summary of Shire data on file.

Synopsis

Anagrelide is extensively metabolised in man to two major metabolites; 3-hydroxy anagrelide (also known as SPD604, BCH24426 or 3-HA) 6,7-dichloro-3-hydroxy-1,5-dihydro-imidazo[2,1-*b*]quinazolin-2-one and RL603, 2-amino-5, 6-dichloro-3, 4-dihydroquinazoline. Earlier pharmacokinetic studies in volunteers have recently been followed by a study in patients with essential thrombocythemia (ET) or other myleproliferative diseases where a notable difference was observed. Patients had much greater exposure (>2 fold) to 3-HA than volunteers possibly due to a longer half-life. 3-HA was found to be the major circulating component in blood representing ~45% of all drug related products in the plasma. The other metabolite RL603 constituted ~33% of the plasma components in these patients. Anagrelide itself represented ~20% of the plasma constituents. Anagrelide had the shortest half-life of 1.7h, followed by 3-HA with a half-life of 3.9 and finally RL603 with a half-life of 8.7h.

Pharmacological evaluation of anagrelide and its metabolites showed that, 3-HA had a comparable inhibitory effect to the parent drug on megakaryocytopoiesis - and potentially therefore platelet formation - while RL603 was inactive. Anagrelide and 3-HA were also found to be inhibitors of PDEIII although 3-HA was almost forty times more potent than the parent drug while RL603 was again virtually inactive. *In vivo* studies in dogs showed that this PDEIII inhibitory activity translated into the expected cardiovascular effects resulting in lowered blood pressure, increased heart rate and increased force of cardiac contraction. Additionally, PDEIII inhibition in the blood platelets resulted in an anti-aggregatory effect with the metabolite being at least 12-13 times more potent than anagrelide.

These data together with the clinical pharmacokinetics are summarised in the in-text table below:-

Compound	*% Total plasma exposure in ET patients*	Plasma t½	Inhibition of megakaryocytopoiesis (IC_{50})	Inhibition of phosphodiesterase III (IC_{50})
Anagrelide	~20%	1.7h	27+/- 10 nM	32nM
BCH24426(3HA)	~45%	3.9h	48+/- 13 nM	0.9nM
RL603	~33%	8.7h	Inactive	40,000nM

In view of the significantly greater exposure to 3-HA than to the drug and considering their relative pharmacological potencies, it is probable that this metabolite contributes

most of the platelet-lowering activity and almost all of the cardiovascular side effects seen in patients treated with anagrelide. Any anti-aggregatory effects would also most likely be due to 3-HA.

Thus the role of 3-HA would appear central to the activity of anagrelide. This contention is supported by the results of an exploration of possible pharmacokinetic–pharmacodynamic relationships in ET patients. This showed a good correlation between chronic exposure to the *active metabolite* and the magnitude of platelet lowering as well as with heart rate but a weaker correlation for the drug itself with respect to platelet lowering and no correlation at all with increase in heart rate.

Thus it would appear that the most appropriate barometer of anagrelide's effects would be plasma levels of 3-HA rather than anagrelide itself.

1. Pharmacokinetics of anagrelide and its metabolites

1.1 Disposition in healthy volunteers

Data from 38 healthy volunteers (age 21-70yrs of age - mean 52) who participated in three separate clinical pharmacokinetic studies on the drug has provided the basis for an overview of pharmacokinetics of anagrelide and its two major metabolites 3-hydroxy anagrelide (otherwise known as BCH24426, SPD604 or 3-HA) 6,7-dichloro-3-hydroxy-1,5-dihydro-imidazo[2,1-*b*]quinazolin-2-one and RL603, 2-amino-5,6-dichloro-3,4-dihydroquinazoline. All subjects ingested a 1mg dose of anagrelide following an overnight fast.

A tabulation of the mean pharmacokinetic parameters is presented in Table 1 while the comparative pharmacokinetic profile of anagrelide and its two metabolites is shown in Figure1.

T_{max} (mean ± relative standard deviation) for anagrelide was 1.3 hours ± 53.8% indicating rapid absorption of the drug. The mean C_{max} value was 4.99 ng/mL ± 74.4% while exposure, in terms of $AUC_{0\text{-}inf}$ was 11.1 ng·h/mL ± 37.6%. Elimination proceeded rapidly in a mono-exponential manner with a mean half-life of 1.5 hours ± 49.8%.

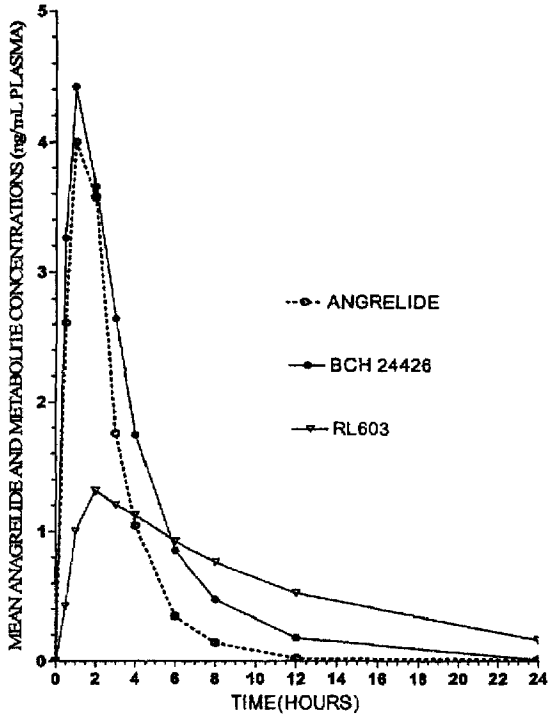
With respect to the kinetics of the active metabolite, 3-HA, the overall mean C_{max} was slightly higher than that for the drug at 5.47ng/ml ± 56.9% and was achieved at a T_{max} of 1.28 hours ± 58.1%. Exposure amounted to 18.0 ng·h/mL ± 35.6 %, some 60% greater than that for the drug itself. The mean half-life of elimination was 2.5 hours ± 28.7%.

For the inactive metabolite, RL603 the mean T_{max} was reached at 2.5 hours ± 58.5% and the corresponding mean C_{max} was 1.36ng/mL ± 34.0%. The mean $AUC_{0\text{-}inf}$ was 16.0ng·h/mL ± 32.3% and mean half-life of elimination was comparatively long at 7.8 hours ± 31.1%.

Attachment G Table 1: Summary of mean pharmacokinetic parameters of anagrelide and BCH24426 from 38 volunteers given a single 1mg dose of Agrylin

Compound	$AUC_{0\text{-}inf}$ ± RSD (%) (ng·h/mL)	C_{max} ± RSD (%) (ng/mL)	T_{max} ± RSD (%) (h)	$t_{1/2}$ ± RSD (%) (h)[1]
Anagrelide	11.1 ± 37.6	4.99 ± 74.4	1.3 ± 53.8	1.5 ± 49.8
BCH24426 (3-HA)	18.0 ± 35.6	5.47 ± 56.9	1.28 ± 58.1	2.5 ± 28.7
RL603	16.0 ± 32.3	1.36 ± 34.0	2.5 ± 58.5	7.8 ± 31.1

Attachment G Figure 1: Mean plasma concentrations of anagrelide, 3-hydroxy anagrelide and RL603 in volunteers



These data presented graphically in Figure 1, show the somewhat greater exposure to 3-hydroxy anagrelide than to the parent drug. Although the mean C_{max} values did not differ that markedly being 5.47ng ng/ml and 4.99/ml for 3-hydroxy anagrelide (BCH24426) and anagrelide respectively, the ratio of metabolite to drug exposure (AUC) was 1.6 :1.

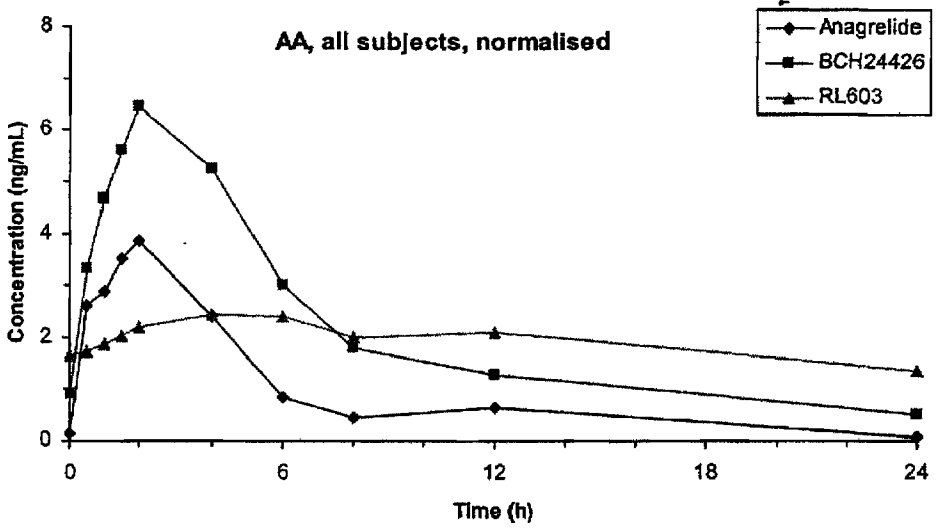
1.2 Disposition in patients with essential thrombocythemia.

The pharmacokinetics of anagrelide and its metabolites were recently investigated in patients with essential thrombocythemia (ET). This study involved a comparison of paediatric patient group (<15years of age) with a more representative adult group of patients (16 –86 years of age – mean 63). Patients were being treated with a variety of different dose levels. Consequently the derived pharmacokinetic parameters were normalised to a 1mg dose and body weight to 70kg. Data from the more representative adult patient group are presented in the Table 3 and Figure 2 below.

Attachment G Table 3: Summary of mean pharmacokinetic parameters of anagrelide and BCH24426 in adult patient group (n=18) at steady state (data normalised to 1mg dose and 70kg body weight)

Compound	$AUC_{0\text{-}tau}$ ±RSD (ng·h/mL)	C_{max} ±RSD (ng/mL)	T_{max} ±RSD (h)	$t_{1/2}$ ±RSD (h)
Anagrelide	19.46 ± 67%	6.22 ± 62%	2.0 ± 68%	1.7± 47%
3-HA (BCH24426)	44.06 ± 40%	8.74 ± 37%	2.3 ± 54%	3.87± 44%
RL603	32.07± 102%	3.20 ± 71.4%	3.8 ± 77.9%	8.69 ± 55.3%

Attachment G Figure 2: Mean plasma concentrations of anagrelide, 3-hydroxy anagrelide and RL603 in ET patients at steady state



1.3 Comparison of patient and volunteer PK data

The pharmacokinetic parameters observed in ET patients do, in several respects, show similarity with those data generated in volunteers. Absorption was comparably rapid with a T_{max} at 2h versus 1.3h seen in volunteers. Peak plasma concentrations of anagrelide and its subsequent half- life of elimination were reasonably similar being 6.22 and 4.99 ng/ml and 1.5 and 1.7h in patients and volunteers respectively.

However the most notable difference was seen in the pharmacokinetics of the active metabolite. In the patient group the mean C_{max} value was 8.74 ± 37% ng/ml compared to 5.47 ± 57% ng/ml in volunteers but the most profound difference was in the exposure being 44.06 ± 40.5% ng·h/mL in patients compared to 18.0 ± 35.6% ng·h/mL in volunteers. Interestingly the half-life of elimination of 3-HA

was longer in patients than in volunteers being 3.9h compared to 2.5h respectively which could account for this greater exposure. This difference is unlikely to be simply due to age since in this respect the subjects were quite similar, with the volunteers ages ranging from 21-70yrs, mean 52 compared to the patients, 16 - 86yrs, mean 63.

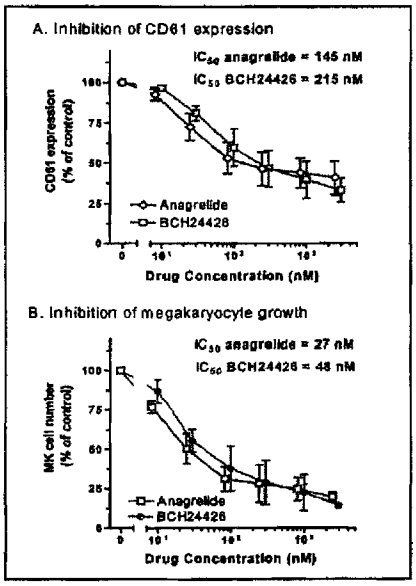
3. Pharmacology of anagrelide and its metabolites

3.1 Primary pharmacology - <u>in vitro</u> screening for platelet lowering potential

The effects of anagrelide and its metabolites on the differentiation of human $CD34^+$ stem cells to megakaryocytes was assessed using a well established model of megakaryocytopoeisis (Cohen-Solal 1997, Cramer 1997). RL603 was found to be inactive in this model (Erusalimsky, Hong & Franklin 2002) despite earlier reports to the contrary (Lane et al 2001). Consequently it is now considered unlikely that this compound (RL603) contributes to the drug products therapeutic effects.

By contrast, 3-hydroxy anagrelide was found to have a comparable IC_{50} value to the parent drug (anagrelide) in affecting the extent of megakaryocyte growth and differentiation (see Figure 3 below). The most marked effect, like anagrelide, was on cell growth. The mean results from several studies showed anagrelide and 3-hydroxy anagrelide to be comparable in their potency to inhibit megakaryocyte growth and differentiation (and ultimately therefore blood platelet formation), having mean IC_{50} values for the former process of 27nM and 48nM respectively.

Attachment G Figure 3: Effects of anagrelide and its metabolite BCH24426 (3-HA) on megakaryocytopoeisis



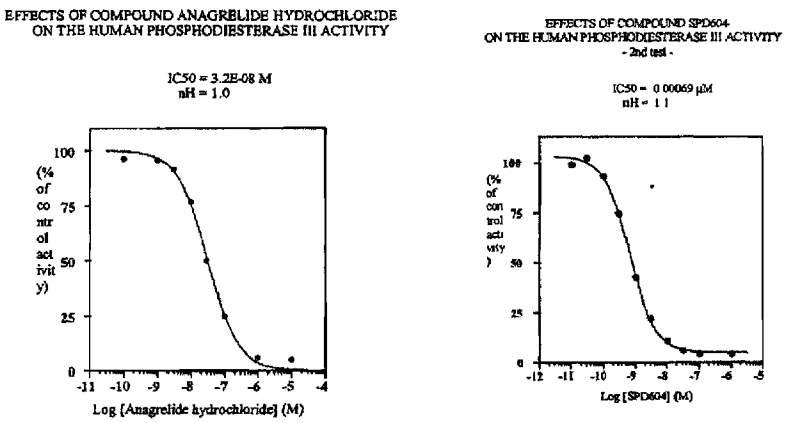
On this basis it is probable that 3-hydroxy anagrelide would make a substantial contribution to the platelet lowering effects of anagrelide especially in view of the clinically observed greater exposure (> 2 fold) to this metabolite than to anagrelide in ET/MPD patients. Furthermore while it is difficult to confidently extrapolate *in vitro* data to the *in vivo* clinical setting due to factors such as plasma protein binding* it is interesting to note that the IC_{50} values for anagrelide and 3-hydroxy anagrelide - equivalent to 7 and 13ng/ml respectively - are of the same order the observed maximum plasma concentrations of anagrelide and its active metabolite seen in patients treated with the drug product.

*The *in vitro* assay examining the effects of compounds on megakaryocytopoeisis contained 10% umbilical cord blood plasma which would effectively account for any inherent differences in plasma protein binding.

3.2 Secondary pharmacology - consequences of inhibition of PDEIII

As anagrelide was already known to inhibit PDEIII, the activity of the metabolites was examined using enzyme derived from human platelets. The active metabolite, 3-hydroxy anagrelide was found to be nearly 40 times more potent than anagrelide itself having an IC_{50} value of ~0.7nM (see Figure 4) This was verified in a second study resulting in average IC_{50} of 0.9nM. By contrast RL603 was essentially inactive. Again while acknowledging the potential pitfalls in *in vitro* - *in vivo* extrapolations it is interesting to note that the clinically observed C_{max}(~9ng/ml) for BCH24426 was approximately 33 fold higher than its *in vitro* IC_{50} for inhibition of PDEIII. By comparison, the C_{max} concentrations for anagrelide would amount to only about three quarters the IC_{50} for its inhibition of PDEIII. The other metabolite of anagrelide, RL603, was an extremely weak inhibitor of PDEIII with an IC_{50} as high as 40,000nM.

Attachment G Figure 4: Effects of anagrelide and 3-HA (SPD604) on human PDEIII



The expected *in vivo* cardiovascular consequences of the PDEIII inhibition such as positive inotropic, chronotropic and vasodilatory activity have been reported previously for anagrelide itself. Recent studies have focussed on examining 3-hydroxy anagrelide in the anaesthetised dog model comparing it to the reference PDEIII inhibitor milrinone. These studies showed 3-HA to have a qualitatively similar cardiovascular profile to milrinone although it was at least 10-20 times more potent. In view of the very much greater potency of 3-hydroxy anagrelide to anagelide in this respect - and the greater plasma exposure seen clinically (metabolite to drug ratio being 2.3:1 - it would seem likely that this metabolite is the major contributor to the observed cardiovascular side effects of the drug product seen in man.

The other anticipated effect of inhibition of PDEIII namely platelet anti-aggregatory activity, was demonstrated for both anagrelide and 3-hydroxy anagrelide (although not evident with RL603). Anagrelide inhibited collagen induced platelet aggregation in human platelet enriched plasma but this effect was only substantial at relatively high concentrations of ~250ng/ml (much higher than would be observed clinically where the usual C_{max} is ~6ng/ml). By contrast 3-HA was much more potent having an IC_{50} of 0.053uM (14ng/ml) within the range encountered clinically (4-16ng/ml after a 1mg dose). RL603 had no effect on aggregation at concentrations up to 1000ng/ml, well above those encountered clinically.

4. Clinical pharmacokinetic – pharmacodynamic correlations.

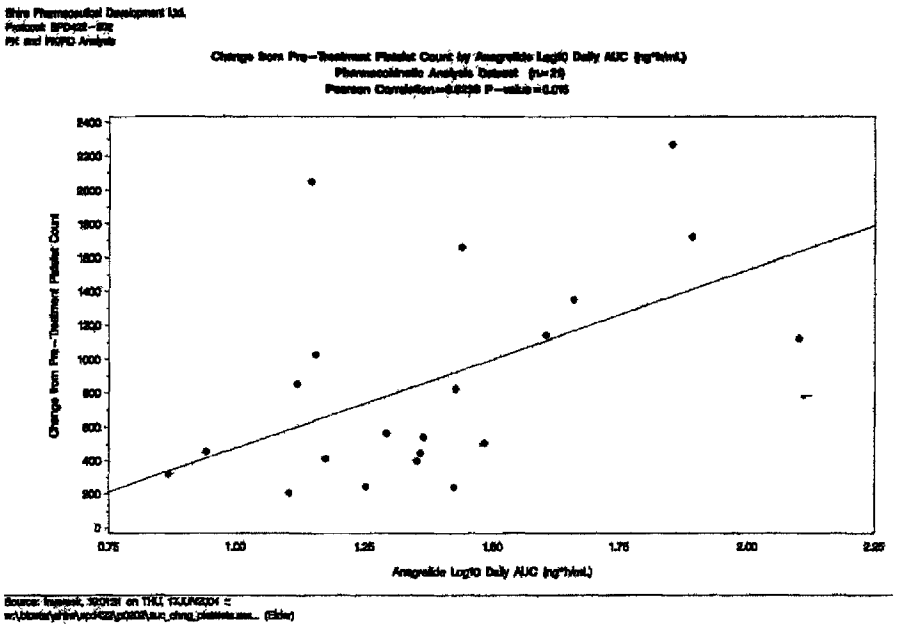
The possibility of a correlation existing between plasma anagrelide and/or active metabolite concentrations and the drug's clinical effects was explored in a group of ET patients being treated with anagrelide. Since the thrombocytopenic effects of anagrelide are only seen following extended multiple dosing, the possible relationship between *chronic* exposure to anagrelide and metabolite (AUC) and change in platelet count (over original baseline values) was examined in a log-linear model. Since the cardiovascular side effects of the drug product such as tachycardia occur acutely this was thought to be more likely related to C_{max} than total exposure. Hence an exploration of the relationship between these levels and heart rate was made for both anagrelide and active metabolite again using a log-linear model.

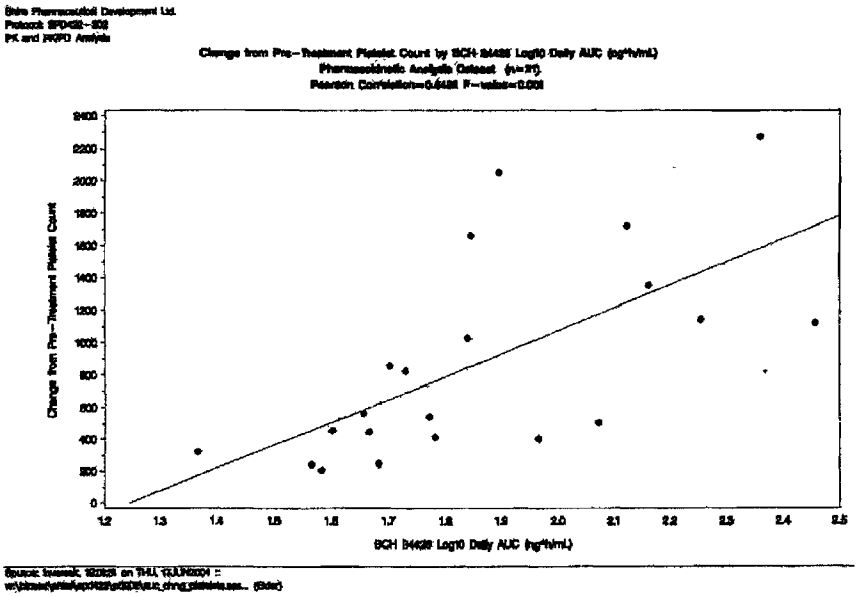
The results of the correlation analysis with platelet count are presented in Table 4 and Figure 5 below. The results show a stronger correlation between log plasma concentrations of the metabolite and changes in platelet count than for anagrelide itself.

Attachment G Table 4: Correlation coefficients for log AUC at steady state and change in platelet count in ET patients

Compound & correlation analysis	N	Pearson correlation coefficient	Probability value
Log anagrelide AUC/ platelet count	21	0.5236	0.015
Log BCH24426 AUC/ platelet count	21	0.6492	0.001

Attachment G Figure 5: Relationship between log anagrelide or 3-hydroxy anagrelide (BCH24426) exposure (AUC) and change in platelet count at steady state in ET patients



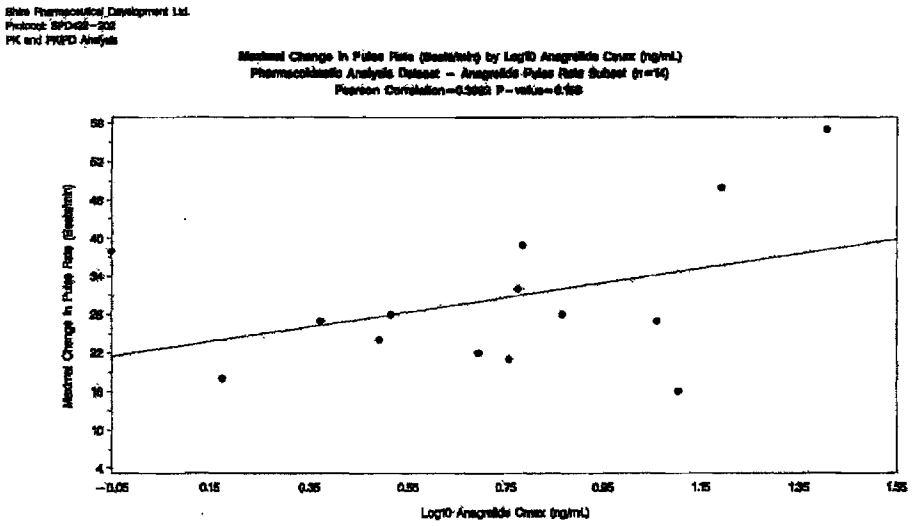


Examination of the correlation with pulse rate showed no correlation between anagrelide C_{max} and pulse rate but an excellent correlation for the active metabolite and pulse rate as shown in Table 5 and Figures 6a and b.

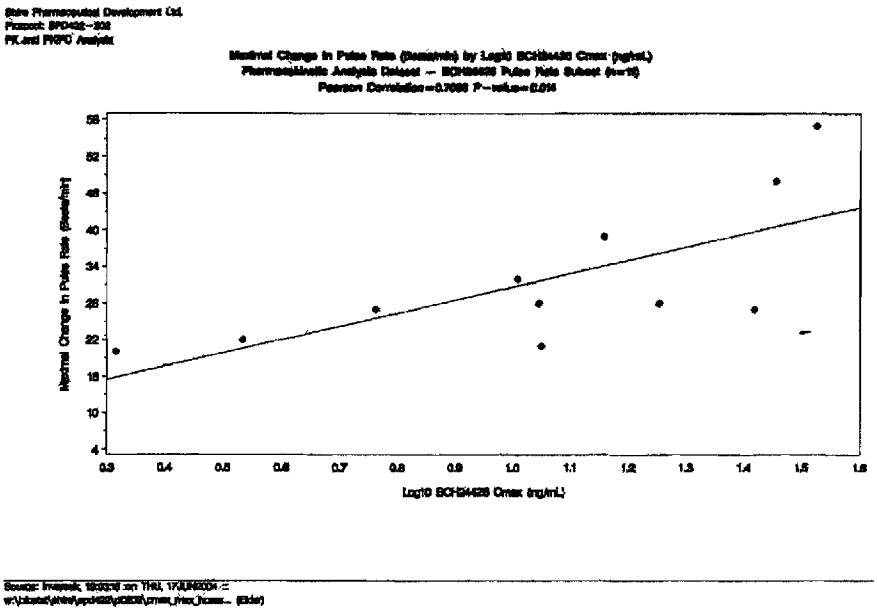
Attachment G Table 5: Correlation coefficients for log Cmax and pulse rate

Compound & correlation analysis	N	Pearson correlation coefficient	Probability value
Log Anagrelide C_{max}/ pulse rate	14	0.3962	0.158
Log BCH24426 C_{max}/pulse rate	11	0.7096	0.014

Attachment G Figure 6a: Relationship between log of anagrelide exposure (AUC) and change in pulse rate at steady state in ET patients



Attachment G Figure 6b: Relationship between log 3-hydroxy anagrelide (BCH24426) exposure (AUC) and change in pulse rate at steady state in ET patients



These correlations are entirely consistent with the proposed relative importance of anagrelide and 3HA in the drug products pharmacological activity.

5. Overall conclusions from the clinical pharmacokinetic and pharmacology studies on the active metabolite's (3-hydroxy anagrelide) role in the drug product activity.

A clinical study in patients has shown 3-hydroxy anagrelide to be the major entity circulating in the plasma of subjects with essential thrombocythemia or other MPDs. *In vitro* studies on the effects of this metabolite on the process of megakaryocytopoeisis suggest that it will have comparable platelet lowering activity to that seen for anagrelide since the compounds appear to be of a similar *in vitro* potency. By contrast, the other metabolite of anagrelide, RL603, is inactive. The predictability of this extrapolation from an *in vitro* screen into *in vivo* platelet lowering activity is supported by data on other compounds. For example hydroxyurea has been shown to inhibit megakaryocytopoeisis in this cell culture model, effects that are known to translate into clinical activity in terms of platelet lowering. In attempting to

transpose this *in vitro* activity into the *in vivo* setting differential factors affecting local availability of the compounds at the site of action (bone marrow) could influence the observed clinical activity e.g. plasma protein binding. However the relative activity of anagrelide and its active metabolite, 3-hydroxy anagrelide, is likely to be maintained since the assay for megakaryocytopoeisis included 10% umbilical cord blood plasma. Thus it might be reasonably assumed that the relative activity *in vitro* may be reflected by comparability *in vivo* . Since the active metabolite accounts for approximately 70% of the therapeutically active entities circulating in blood then given this comparability of potency with anagrelide it is likely to account for most of the platelet lowering activity of the drug product Agrylin. That 3-hydroxy probably plays such a major role in the platelet reducing properties of the drug product is reflected by the stronger PK-PD correlation for this compound rather than for anagrelide itself.

With respect to the contribution the metabolites make to the observed cardiovascular side effects of anagrelide (tachycardia and palpitations), RL603 being essentially inactive as a PDEIII inhibitor is unlikely to play any role but 3-hydroxy anagrelide which is 40 times more potent than anagrelide itself as an inhibitor of PDEIII will be the dominant contributor. Once again however the question might arise as to the predictability of the *in vivo* cardiovascular (CVS) effects from the *in vitro* inhibition of PDEIII. The consequences of this inhibition would be to increase levels of cyclic AMP, a second messenger, resulting in increased myocardial contractility and vasodilation in the venous and arterial circulation. There are many examples of drugs where this *in vitro* inhibition of PDEIII has been shown to predictably translate into CVS effects in man e.g. milrinone, amrinone enoximone, olprinone etc (Movsesian 2003). Once again however differential anagrelide and metabolite availability at the site of action - here the myocardium - could be determined by factors such as differential non-specific tissue and/or plasma protein binding. However the magnitude of the difference in the respective potencies to inhibit PDEIII are so large that such differences would most likely to be preserved *in vivo*. Finally investigation of the PK-PD between pulse rate and anagrelide or 3-HA C_{max} showed evidence only for such a relationship with the metabolite.

In summary the investigations carried out consistently indicate that the most appropriate barometer for assessing Agrylin's biological effects would be plasma levels of 3-hdyroxy anagrelide rather than those of the parent drug.

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H

ATTACHMENT H

Evidence for pre-systemic metabolism of anagrelide

This represents a summary of Shire data on file

Summary

The unavailability of a clinical intravenous formulation of anagrelide has precluded the generation of absolute oral bioavailability data on this compound. Nevertheless there is considerable indirect evidence to support the belief that anagrelide undergoes a significant first pass metabolism in the formation of its active metabolite (6,7-dichloro-3-hydroxy-1,5-dihydro-imidazo[2,1-*b*]quinazolin-2-one) known as BCH24426. This comprises:-

• An estimate of the absolute oral bioavailability of the drug in the minipig, a species recently shown for several drugs to give reasonable estimates of human pharmacokinetic parameters, indicated this to be ~20%.

• Following oral administration of anagrelide to volunteers the superimposition of the early plasma concentration-time profiles for drug and active metabolite indicates formation of the metabolite by first pass.

• A study in subjects with moderate hepatic impairment showed an 8-fold increase in anagrelide AUC, a two fold increase in half-life and what appeared to be a four fold increase in Vd/F. While the latter could represent a real change in volume due to disease induced changes in plasma protein binding of the drug, it is possible that this change may be at least in part due to a significant increase in bioavailability. This would imply that bioavailability in the absence of hepatic impairment was low.

• A theoretical estimate of the extent first pass metabolism may be made from the equation:-

$$F = 1/(1 + (CLb/F)/QH$$

Where CLb is blood clearance = CL/F x C/Cb (=1.196) and QH, is the liver blood flow (1.35L/min). The assumptions underlying this equation are that hepatic extraction is the sole cause of reduced bioavailability and that elimination occurs exclusively by hepatic metabolism.

- Evidence for the former assertion comes from two human radiolabelled studies which showed between 72-78% of the administered radiolabel was recovered in the urine implying near quantitative absorption. In the light of this, bioavailability is likely to reflect pre-systemic metabolism.

- Since there is no evidence for luminal degradation of the drug from biotransformation by gut flora or human intestinal microsomes it may be assumed that pre-systemic metabolism is due to hepatic biotransformation. There is no evidence of significant renal clearance of the drug itself.

Individual estimates of bioavailability from 69 volunteers, using Eq. 1, indicated a mean ± SD of 43.4% ± 9.8. Data from a smaller group of 16 essential thrombocythemia patients indicated a mean ± SD bioavailability of 52.6% ± 12.5. Thus between 47-57% of the drug is removed by hepatic extraction during the initial passage through the liver.

• Taken together, all these data suggest anagrelide undergoes significant first pass metabolism in the formation of its active metabolite,3-hydroxy anagrelide.

1. Background

Anagrelide (imidazo [2,1-b] quinazolin- 2(3H)-one, 6,7-dichloro-1,5-dihydro, monohydrochloride), is extensively metabolised in man to two major metabolites; 3-hydroxy anagrelide (6,7-dichloro-3-hydroxy-1,5-dihydro-imidazo[2,1-*b*]quinazolin-2-one, also known as SPD604, BCH24426 or 3-HA) and a subsequent biotransformation product RL603, 2-amino-5,6-dichloro-3,4-dihydroquinazoline. 3-hydroxy anagrelide is equipotent with the parent dug in its *in vitro* effects on megakaryocytopoesis and therefore potentially platelet lowering but 40 times more potent as a PDEIII inhibitor and therefore as an inotrope, chronotrope and vasodilator. The further metabolite, RL603, is essentially inactive in these screens (Erusalimsky, Hong, and Franklin 2002)

It is believed that the primary active metabolite of anagrelide, 3-hydroxy anagrelide, is formed extensively during first pass through the liver although there are no absolute oral bioavailability data to confirm this assertion. Since essential thrombocythemia is a chronic condition marked by an elevation of blood platelets, a clinical intravenous formulation was not required and thus never developed. This has regrettably precluded the *direct* measurement of absolute oral bioavailability although other data provide a valuable insight into the likelihood of pre-systemic metabolism of anagrelide.

2. Non-clinical evidence for first pass metabolism

2.1 Oral bioavailability data in the minipig

Recently a number of reviews appear to suggest that the minipig may be a good model for man in terms of drug absorption and disposition. This has been attributed to the similarity of its cytochromes P450 and its gastrointestinal tract to those in man (Anzenbacher et al 1998).

Data have been generated in a non-crossover design in two groups of three female Göttingen SPF minpigs. On the first dosing occasion each of three animals was given 2 x 0.5mg Agrylin® capsules with 20 mL water after an overnight fast. Fasting was continued for at least a further 4 hours. On a further dosing occasion an additional three minipigs were given a 1mg intravenous bolus of anagrelide in 2mL of propylene glycol, again after an overnight fast. In both cases heparinised blood samples were drawn at various times for assessment of plasma drug concentrations using a validated LC/MS-MS method.

Comparison of the oral AUC_{24h} (10.3ng.h/mL) with the intravenous AUC_{inf} estimate (50.7ng.h/mL) suggested an oral bioavailability for anagrelide in minipigs of about 20%. However this estimate should be treated with caution, since it is based on data from only two of the three animals given the intravenous dose (data for the third animal were lost) and due to a failure to be able to compare AUC over the same interval i.e., infinity. Nevertheless it does give an indication of a potentially low absolute bioavailability of anagrelide.

Attachment H Table 1: Pharmacokinetic parameters for anagrelide in Göttingen minipigs given 1mg of the drug orally or intravenously

Dosing	Animal No.	Tmax	Cmax	AUC24h
		(h)	(ng/mL)	(ng.h/mL)
	1	3.0	1.29	8.3
oral	2	8.0	1.09	10.4
	3	3.0	1.98	12.3
	Mean	4.7	1.45	10.3
	(CV%)	(62)	(32)	(20)
	4	0.0	83.5	42.7*
iv	5	0.0	90.6	58.7*
	Average	0.0	87.1	50.7*

* $AUC_{infin.}$

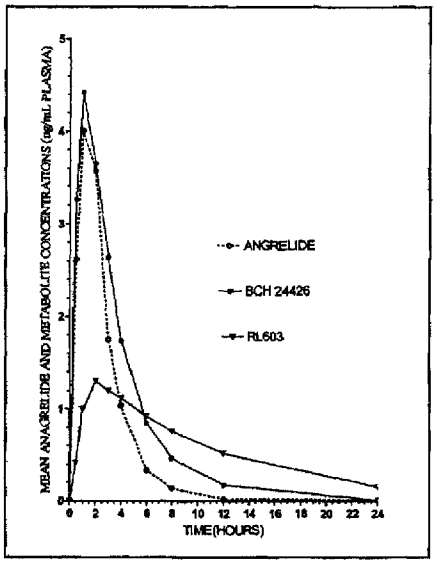
3. Clinical evidence for first pass metabolism

3.1 Superimposability of the rate of formation of the metabolite with absorption of the drug

Data from 38 healthy volunteers age (range 21-76 mean 52 yrs) who participated in three separate clinical pharmacokinetic studies showed that following a 1mg oral dose of the drug under fasting conditions the drug was rapidly absorbed (T_{max} of 1.3 hours) and eliminated (terminal half-life of 1.5 h).

Most importantly the rate of formation of the active metabolite 3-hydroxy anagrelide appeared to proceed in parallel with the absorption of the drug (Figure 1). The concentration –time profiles were superimposed suggesting the metabolite was formed during the first pass of anagrelide through the liver.

Attachment H Figure 1: Plasma concentration time profile for anagrelide and its active metabolite, 3-hydroxy anagrelide, in man following a single 1 mg oral dose of the drug under fasting conditions



3.2 Evidence from hepatic impairment PK study

A pharmacokinetic study in 10 subjects with moderate hepatic impairment (see Table 2) given a single 1mg oral dose of anagrelide revealed an 8-fold increase in total exposure (AUC) to anagrelide. Part of this increase may be related to the 2.2-fold increase in half-life. Surprisingly however, the apparent volume of distribution, (V_z/F), decreased by almost 4-fold. While this could be due to increased plasma protein binding (for example brought about by an increase in α_1-acid glycoprotein in these hepatically compromised subjects, assuming this to be the binding protein), other mechanisms may play a role. For example, an increase in bioavailability may occur,

as is frequently the case for drugs exhibiting extensive first pass hepatic metabolism. This observation may also provide supporting evidence for first pass metabolism of anagrelide.

Attachment H Table 2: Anagrelide PK parameters in hepatic impairment [arithmetic means (SD)*]

Parameter	Hepatically Impaired	Healthy
C_{max} (ng/mL)	[N=10] 13.2 (8.12)	[N=10] 6.09 (6.59)
t_{max} (hr)	[N=10] 2.00 (0.50-2.00)	[N=10] 1.00 (1.00-3.00)
$AUC_{0-\infty}$ (ng·hr/mL)	[N=9] 83.8 (67.0)	[N=9] 10.8 (6.03)
$t_{1/2z}$ (hr)	[N=9] 3.30 (1.40-10.1)	[N=9] 1.53 (1.00-5.20)
V_z/F (L)	[N=9] 87.9 (32.4)	[N=9] 324 (250)
CL_T/F (L/hr)	[N=9] 26.0 (23.5)	[N=9] 119 (57.7)
CL_R (L/hr)	[N=0]	[N=5] 0.0094 (0.00531)

*except tmax which is median & range .

3.3 Equation derived estimates of bioavailability

In attempting to estimate the extent of first pass metabolism of anagrelide, the following equation was applied:-

$F = 1/(1 + (CL_b/F)/QH)$ (Rowland and Tozer 1995)

Where QH the liver blood flow (assumed to be 1.35L/min) The relationship between blood clearance and plasma clearance is given by $CL_b/F = CL/F \times C/C_b$ (the ratio $C/C_b = 1.196$,)

Use of this equation makes the following assumptions:-

- Hepatic extraction is the only cause of reduced bioavailability i.e. no issue of incomplete absorption or degradation of the drug in the gut lumen or wall.
- No extrahepatic elimination of the drug e.g. no renal clearance of the drug.

Evidence for hepatic extraction as the sole cause of reduced bioavailability

(i) Urinary recovery of orally administered radiolabelled drug showing good absorption. Two human radiolabelled studies showed extensive renal excretion of orally administered radioactivity. Assuming anagrelide is not degraded in the gut this

would suggest extensive absorption of anagrelide (Gaver et al 1981). The results of these studies are presented in Table 3.

Attahcment H Table 3: Recovery of radioactivity in volunteers given a single oral dose of 1mg [^{14}C]- anagrelide

Study number & subject number	% administered dose recovered in urine	% administered dose recovered in faces	Grand totals
13,970-107A/1	80.3	15.4	95.7
13,970-107A /2	78.9	17.7	96.5
13,970-107A /3	78.7	23.3	102
13,970-107A /4	77.0	22.0	99.0
13,970-107A /5	76.8	22.7	99.5
Means ± SD	**78.34 ± 1.5**	**20.2 ± 3.5**	**98.5 ± 2.5**
3/1774/1	80.0	9.3	89.3
3/1774/2	71.2	2.9	74.1
3/1774/3	60.7	14.2	74.9
3/1774/4	73.3	17.7	91.0
3/1774/5	74.0	7.1	81.1
Means ± SD	**71.8 ± 7.0**	**10.2 ± 5.8**	**82.1 ± 7.9**
***Overall mean* ± SD**	**75.1 ± 5.9**	**15.2 ± 6.9**	**90.3 ± 10.3**

13,970/017A study used a seven day collection period
3/1774 study (Gaver et al 1981) used a six-day collection period

Other volunteer studies showed rapid absorption from the Agrylin capsule formulation (see Figure 1). Across three separate volunteer studies involving the administration of 1mg of the drug to 38 fasted subjects, T_{max} was achieved with mean ± CV of 1.3h ± 53.8%. Since rapid absorption is usually associated with complete absorption these results suggest quantitative absorption of anagrelide.

(ii) Evidence for lack of degradation in gastrointestinal tract and gut wall
An investigation of the possible *in vitro* biotransformation of [^{14}C] anagrelide by *human gut microflora* was conducted using a human faecal preparation (bacterial cell suspension). After incubation of this preparation with a 6 µM solution of the radiolabelled anagrelide for up to 24 hours under anaerobic conditions, subsequent radio-HPLC of the incubates revealed no convincing evidence of bioconversion. Less than 10% of the drug had apparently disappeared, but this was not significantly different from the control incubates, conducted in the absence of bacterial cell suspension. On the basis of these results it was concluded that there was no evidence for gut luminal metabolism of anagrelide.

An investigation of the potential for metabolism of anagrelide by the gut wall was undertaken using *human intestinal microsomes*. Following a 45 minute incubation period the positive control compound, midazolam, was near completely metabolised but 86.4% of anagrelide was still remaining comparable to the NADPH deficient control incubations (see Table 4).

Attachment H Table 4: Investigation of *in vitro* metabolism of anagrelide by use of human intestinal microsomes

	% Parent Compound Remaining					
Compound ID	0 min	5 min	15 min	30 min	45 min	Minus NADPH control
Anagrelide	100.0	98.3	92.7	83.1	86.4	79.3
Midazolam	100.0	66.1	27.9	5.9	2.1	100.7

(The apparent small reduction in substrate concentrations was attributed to non-specific binding of anagrelide)

It was therefore concluded that anagrelide was not significantly metabolised by human intestinal microsomes.

Direct evidence of hepatic metabolism

Direct evidence for the *hepatic metabolism* of anagrelide comes from work with human hepatocytes. [^{14}C]-anagrelide (2µM) was incubated with cryopreserved human hepatocytes in Williams' Medium E (supplemented with HEPES (10 mM), foetal calf serum (10%, v/v), dexamethasone (1 µM) and SPITE (1%, v/v) for 3h. Samples were periodically removed and submitted to radio-HPLC for quantification of disappearance of drug and appearance of metabolites. The results showed a time dependent decrease in the anagrelide concentrations in the incubates and a concomitant increase in the amount of the two metabolites BCH24426 and RL603.

Subsequent work with expressed cytochromes (Supersomes®) showed that amongst the isoforms evaluated (3A4, 2C9, 2C19, 2D6, and 1A2) only CYP1A2 appeared to metabolise anagrelide. Since this enzyme is known to be present in the liver but largely absent in the gut wall this provides further support for the assertion of hepatic metabolism of anagrelide.

Evidence for lack of renal clearance of anagrelide

No significant amount of unchanged anagrelide has been found in the urine of any of the volunteers involved in the human radiolabelled study (estimated <1%, Gaver et al 1981). A more formal study in which a quantitative bioanalytical method was applied to the analysis of urine from the ten healthy volunteers age, matched for age, weight,

and sex in a renal impairment study confirmed this initial indication of an essential absence of unchanged drug in the urine (see Table 5).

Attachment H Table 5: Amount of anagrelide excreted in the urine (Ae) in ten volunteers (healthy matched controls from renal impairment study, SPD422-103) after a single 1mg oral dose

Subject number	Ae 0-4h ng	Ae 4-8h ng	Ae 8-12h ng	Ae 12-24h ng	Grand totals	% administered dose
1	183.2	27.75	<0.05	<0.05	210.9	0.021
2	44.8	<0.05	<0.05	<0.05	44.8	0.005
3	75.15	<0.05	<0.05	<0.05	75.15	0.008
4	139.1	<0.05	<0.05	<0.05	139.1	0.014
5	48.96	24.40	<0.05	<0.05	73.36	0.007
6	38.74	<0.05	<0.05	<0.05	38.74	0.004
7	91.8	9.86	<0.05	<0.05	101.7	0.010
8	286.65	19.4	<0.05	<0.05	306.0	0.031
9	161.7	23.7	<0.05	<0.05	185.4	0.019
10	70.0	19.04	<0.05	<0.05	89.04	0.009
Means ± sd	114 ± 79	12.4 ± 11.6	<0.05	<0.05	126.4 ± 84.8	0.013 ± 0.01

3.4 Derivation of estimates of oral bioavailability of the drug

Having satisfied the criteria for use of the equation, F = 1/(1 + (CLb/F)/QH) this was applied to estimating the bioavailability in 69 volunteers used in four clinical PK studies. These volunteers were the "control" subjects i.e. age, weight and sex matched healthy comparators for the renal and hepatic studies while for the food and aspirin interaction studies these data were derived from volunteers who were fasted or who took the drug without aspirin respectively.

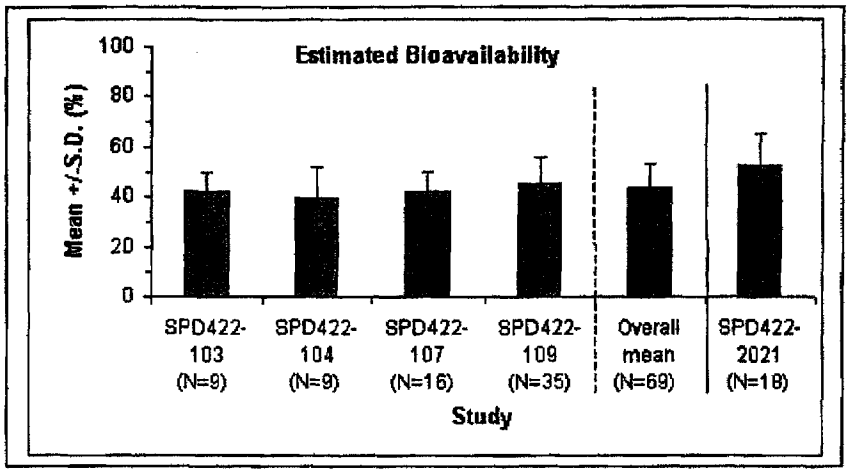
The data for the patient study were drawn from a paediatric vs adult comparison with the latter group (n=18) providing the data used here. The results of these analyses are shown in the Table 6 and Figure 2.

Attachment H Table 6: Estimated oral bioavailability in healthy volunteers and ET/MPD patients

Study	Mean %	S.D.	N
renal impairment study	42.4	7.6	9
hepatic impairment study	39.7	12.4	9
aspirin interaction study	42.1	7.8	16
food interaction study	45.1	10.4	35
Overall mean	43.4	9.8	69
ET patient study[1]	52.6	12.5	18

[1] after repeat administration; normalised to 70kg bodyweight

Attachment H Figure 2: Comparison of estimates of anagrelide's bioavailability in various clinical pharmacokinetic studies in *volunteers and patients*



These estimates of bioavailability - and hence first pass metabolism - are remarkably consistent between all the healthy volunteers with a mean ± S.D. 43.4 %± 9.8. Those data from patients are not dramatically different showing only a slightly higher bioavailability (i.e. lower first pass) but in either case the extent of pre-systemic metabolism ranged from 47-57%.

4. Conclusion

On the basis of all the data presented here it would seem that anagrelide undergoes a major first pass effect of approximately 50% giving rise to its primary active metabolite, BCH24426.

5. References

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EXHIBIT 2

ADIS DRUG EVALUATION

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Anagrelide

A Review of its Use in the Management of Essential Thrombocythaemia

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Data Selection

Sources: Medical literature published in any language since 1980 on anagrelide, identified using MEDLINE and EMBASE, supplemented by AdisBase (a proprietary database of Adis International). Additional references were identified from the reference lists of published articles. Bibliographical information, including contributory unpublished data, was also requested from the company developing the drug.

Search strategy: MEDLINE search terms were 'anagrelide' or 'BL-4162A'. The EMBASE and AdisBase search term was 'anagrelide'. Searches were last updated 19 December 2005.

Selection: Studies in patients with essential thrombocythaemia who received anagrelide. Inclusion of studies was based mainly on the methods section of the trials. When available, large, well controlled trials with appropriate statistical methodology were preferred. Relevant pharmacodynamic and pharmacokinetic data are also included.

Index terms: Anagrelide, essential thrombocythaemia, pharmacodynamics, pharmacokinetics, therapeutic use, tolerability.

Contents

Summary

Abstract

Anagrelide (Agrylin®, Xagrid®) is an oral imidazoquinazoline agent which is indicated in Europe for the reduction of elevated platelet counts in at-risk patients with essential thrombocythaemia who are intolerant of or refractory to their current therapy, and in the US for the reduction of elevated platelet counts and the amelioration of thrombohaemorrhagic events in patients with thrombocythaemia associated with myeloproliferative disorders.

Anagrelide is well established as an effective platelet-lowering agent in most patients with essential thrombocythaemia, including both treatment-naive patients and those refractory to other cytoreductive therapy. Results of the only randomised trial to date (the Primary Thrombocythaemia 1 [PT1] study) indicated that the composite primary endpoint (arterial or venous thrombosis, serious haemorrhage or death from vascular causes) occurred more often in recipients of anagrelide plus aspirin than in those receiving hydroxycarbamide (hydroxyurea) plus aspirin. This trial also indicated that the incidence of the secondary endpoints transient ischaemic attack and gastrointestinal bleeding favoured hydroxycarbamide plus aspirin, while the incidence of venous thrombosis favoured anagrelide plus aspirin. There were no differences between the groups in the incidence of secondary endpoints myocardial infarction, stroke, unstable angina, pulmonary embolism, hepatic-vein thrombosis, other serious haemorrhage or related deaths. The design of the PT1 study has been queried with respect to the heterogeneous nature of the study population (possible inclusion of patients with early myelofibrotic disease) and the concomitant use of aspirin (interaction with anagrelide causing increased bleeding events). Further data are therefore required before the role of anagrelide in essential thrombocythaemia can be finalised. In the meantime, when considering treatment options for patients with this disorder, anagrelide's positive effects on platelet function, lack of mutagenicity and lack of association with leukaemia or angiogenesis must be balanced against its comparative expense and positive inotropic effects. Thus, the role of anagrelide in the management of high-risk patients with essential thrombocythaemia will ultimately depend on individual patient assessment and future clarification of the potential leukaemogenicity of hydroxycarbamide.

Pharmacological Properties

Anagrelide and its active metabolite 3-hydroxy anagrelide specifically, reversibly and dose-dependently block the maturation of late-stage megakaryocytes, thus reducing platelet counts in patients with essential thrombocythaemia. The drug appears to normalise platelet coagulant and endothelial function, does not stimulate myelofibrotic progression and, unlike hydroxycarbamide, is not associated with angiogenesis or damage to DNA. The inhibitory effect of anagrelide, and particularly of the 40-fold more potent 3-hydroxy anagrelide, on phosphodiesterase III results in positive inotropic effects (vasodilation, and increased heart rate and contractility) and potential for pharmacodynamic interactions with other phosphodiesterase inhibitors.

The pharmacokinetics of anagrelide are linear in the 0.5–2mg dose range. After oral administration, anagrelide is rapidly absorbed and the drug is metabolised, mainly during first pass, to two main metabolites, the active 3-hydroxy anagrelide and the inactive 5,6-dichloro-3,4-dihydroquinazol-2-ylamine. Peak plasma concentrations of anagrelide and the active metabolite are reached in about 2 hours. Systemic exposure to 3-hydroxy anagrelide is about twice that of the parent compound in patients with essential thrombocythaemia. The terminal half-lives of anagrelide and the active metabolite are 1.7 and 3.9 hours in this patient group. Since metabolism occurs primarily via cytochrome P450 1A2, interactions with drugs such as fluvoxamine are possible. There is no evidence of an interaction with hydroxycarbamide, digoxin or warfarin. Individual titration of dosages allows the effects of age or hepatic or renal impairment to be taken into consideration.

Therapeutic Efficacy

Early noncomparative trials have clearly indicated that anagrelide lowers platelet counts in most (complete plus partial response in 82–98%; complete response in 38–88%) patients with essential thrombocythaemia. Complete response rates of approximately 50% were seen in patients who were unresponsive to previous hydroxycarbamide treatment in two small trials. Clinically significant reductions usually occurred well within the first month of treatment. Anagrelide and hydroxycarbamide (each with concomitant aspirin) reduced platelet counts to a similar extent after 9 months' treatment in the randomised nonblind PT1 trial in high-risk patients with essential thrombocythaemia, but hydroxycarbamide appeared to act more quickly.

Previous experience of arterial or venous thrombosis or haemorrhage was relatively high at baseline in the PT1 trial. The overall composite primary endpoint of serious thrombohaemorrhagic events favoured hydroxycarbamide. Secondary endpoints indicated that while anagrelide plus aspirin was superior to hydroxycarbamide plus aspirin as measured by the lower incidence of deep-vein thrombosis, hydroxycarbamide plus aspirin was superior with respect to the incidence of transient ischaemic attacks and serious gastrointestinal haemorrhage. There were no differences between the groups in the individual incidences of myocardial infarction, stroke, unstable angina pectoris, pulmonary embolism, hepatic-vein thrombosis, other serious haemorrhages or death from thrombosis or haemorrhage. The concomitant administration of aspirin in the PT1 trial is thought to have resulted in a synergistic action resulting in increased bleeding events.

Essential thrombocythaemia was less likely to progress to myelofibrosis in hydroxycarbamide recipients than in anagrelide recipients in the PT1 trial; howev-

er, the baseline risk for myelofibrosis was not taken into account in the diagnostic methodology, with subsequent potential for imbalance of prefibrotic conditions. The incidence of transformation to acute myeloid leukaemia/myelodysplasia or polycythaemia vera and deaths from transformation were similar in the two groups, although follow-up was possibly inadequate for reliable estimation.

Tolerability

The most common adverse events associated with oral anagrelide are headache, palpitations, diarrhoea, asthenia, oedema, nausea, abdominal pain and dizziness. The incidence and severity of many adverse events decreases with extended treatment in some but not all patients. The incidence of serious adverse events is higher in older patients. Cardiovascular effects such as congestive heart failure or arrhythmia are uncommon. Palpitations, headache, noncardiac oedema, diarrhoea and abdominal pain were more common in recipients of anagrelide plus aspirin than in those receiving hydroxycarbamide plus aspirin in the PT1 trial of high-risk patients with essential thrombocythaemia. Dermatological events (mainly leg ulcers) and diabetes mellitus occurred more often with hydroxycarbamide and white-cell counts were persistently lower in this group. There were no differences between the groups in the incidence of other gastrointestinal symptoms, anaemia, cardiac failure, arrhythmia or minor haemorrhage.

1. Introduction

Essential thrombocythaemia is a myeloproliferative disorder which is associated with a sustained increase in the number of platelets in peripheral blood. Recent investigations have indicated that an acquired mutation on the Janus kinase 2 gene (Jak2V617F) is present in many patients with myeloproliferative disorders, including essential thrombocythaemia.[1] Although 50–70% of patients with essential thrombocythaemia have no symptoms at presentation,[2] characteristic symptoms can develop over the course of the disease. These include microvascular complications caused by spontaneous activation and aggregation of hypersensitive thrombocythaemic platelets (peripheral paraesthesia of the extremities, headache, dizziness, visual disturbances, erythromelalgia, transient ischaemic attacks), thromboembolic complications (pulmonary embolism, splenic infarction, arterial or venous thrombosis, angina pectoris, myocardial infarction) and haemorrhagic complications (oral, intracerebral, gastrointestinal or cutaneous bleeding).[3,4]

Essential thrombocythaemia, which current evidence indicates occurs in about 2.5 per 100 000 person-years,[5] is commonly diagnosed according to criteria initially devised by the Polycythaemia Vera Study Group (PVSG).[6] The amended criteria comprise a platelet count $>600 \times 10^9$/L for ≥2 months and no evidence of reactive thrombocytosis, polycythaemia vera, iron deficiency, chronic myeloid leukaemia, myelofibrosis or myelodysplastic syndrome.[7] Since diagnosis is largely a matter of exclusion, the resulting patient cohort with essential thrombocythaemia is likely to be heterogeneous. When bone marrow histopathology is included as a diagnostic feature, as in the WHO criteria for essential thrombocythaemia, patients with prefibrotic or early fibrotic chronic idiopathic myelofibrosis are excluded.[8,9] Patients diagnosed using the PVSG criteria are thus much more likely to progress to overt myelofibrosis.[8] The discovery of the Jak2V617F mutation in patients with myeloproliferative disorders will also have repercussions on the future diagnosis, classification and treatment of essential thrombocythaemia.[1]

Life expectancy does not appear to be affected in patients with essential thrombocythaemia,[10,11] especially in those diagnosed using bone marrow histopathology.[8] Older patients and those having experienced a previous thrombotic event are considered to be high-risk patients.[11,12] The incidences of

major thrombotic and haemorrhagic events in patients with essential thrombocythaemia, the cause of much of the morbidity and mortality associated with the disease, have been estimated at 7–17% and 8–14%, respectively, in retrospective cohort studies using a wide variety of inclusion criteria, clinical settings and event definitions.[13] Major thrombotic complications have been estimated to occur at a rate of 3–4% per patient-year in low-risk patients and 11% per patient-year in high-risk patients.[13] The cause of clonal evolution of essential thrombocythaemia into idiopathic myeloid metaplasia, myelodysplastic syndrome or acute myeloid leukaemia is not yet clear. While some advocate the presence of a small but real risk of delayed transformation occurring as a result of natural disease progression,[5] others suggest that transformation may be caused by cytoreductive therapy or a mixture of both intrinsic and extrinsic factors.[12]

Anagrelide (Agrylin®, Xagrid®)[1] is an oral imidazoquinazoline agent with platelet-lowering activity in humans.[3] This review focuses on the efficacy and tolerability of anagrelide in the treatment of adults with essential thrombocythaemia.

2. Pharmacodynamic Properties

2.1 Effects on Bone Marrow/Platelets

The mechanism of action of anagrelide has not yet been fully characterised. The lack of animal models of the thrombocytopenic effects of anagrelide (which are more evident in humans than in laboratory animals) has limited study of the pharmacodynamic effects of the drug.[14]

Essential thrombocythaemia is associated with hyperproliferation of megakaryocytes, with increased cell size and ploidy (higher proportions of 32N or higher ploidy cells versus healthy controls), and a mean platelet turnover rate about six times higher than that in healthy controls.[15] Anagrelide and its active metabolite 3-hydroxy anagrelide (section 3.1) specifically prevent megakaryocyte differentiation *in vitro*.[16] 3-Hydroxy anagrelide, like the parent compound, has a marked effect on megakaryocyte growth and is similar in potency and specificity to anagrelide in this respect.[16] The development of megakaryocytes was inhibited by 50% at concentrations of 26 and 44 nmol/L by anagrelide and 3-hydroxy anagrelide, respectively.[16] It is likely that 3-hydroxy anagrelide contributes substantially to the clinical platelet-lowering effects of the formulation, considering the higher systemic exposure to the metabolite than to the parent compound (section 3.1).[17]

Anagrelide dose-dependently and reversibly prevents the maturation of megakaryocytes in non-mitotic late stages of development (the size, surface irregularity, optical density and ploidy of the megakaryocytes are reduced),[15,18-20] which results in increased numbers of precursor cells (promegakaryoblasts and megakaryoblasts).[20] The number of megakaryocytes may also be decreased by anagrelide.[15]

Anagrelide does not affect haematopoietic stem cells and does not damage DNA.[21,22] Oral treatment with either anagrelide or hydroxycarbamide (hydroxyurea) resulted in increased incidence of promegakaryoblasts and microforms in 20 patients with initial to early-stage chronic idiopathic myelofibrosis and thrombocythaemia in a bone marrow immunohistochemical and morphometric study.[22] However, while no such changes were seen with anagrelide, hydroxycarbamide was associated with significant abnormalities of megakaryocytic differentiation indicative of dysplastic changes and therefore of leukaemogenic potential.[22]

The platelet turnover rate is reduced close to normal rates but platelet survival is unaffected in patients with essential thrombocythaemia receiving anagrelide.[15,23] Thus, oral anagrelide caused reversible reductions in platelet counts in healthy volunteers[3] and platelet counts were normalised in patients with essential thrombocythaemia.[24,25] This thrombocytopenic effect, however, does not occur in all patients (see section 4).

In contrast to hydroxycarbamide and interferon-α, anagrelide does not appear to be associated with

1 The use of trade names is for product identification purposes only and does not imply endorsement.

bone marrow angiogenesis in patients with essential thrombocythaemia.[20,26] Levels of platelet count-corrected vascular endothelial growth factor (VEGF) and platelet factor 4 (PF4), markers of angiogenesis, were increased at baseline versus those in healthy controls and were normalised with anagrelide therapy; however, levels were increased further in patients receiving hydroxycarbamide or interferon-α (n = 13).[26] Similarly, bone marrow vessel-related CD34+ progenitor cell levels, also used to mark angiogenesis, were not increased by anagrelide (n = 15).[20]

Many abnormal aspects of platelet function associated with essential thrombocythaemia are improved with anagrelide treatment. Platelet count-corrected thromboxane B2 values tended toward normalisation in 17 patients with essential thrombocythaemia during anagrelide-induced remission.[27] Similarly, platelet coagulant activity and endothelial function (PF4, prothrombin fragment 1+2, plasmin/α2-antiplasmin complex, plasminogen activator inhibitor-1, tissue factor pathway inhibitor) were normalised in 17 patients with essential thrombocythaemia who responded to anagrelide.[28] In this and other studies, normalisation of endothelial function was associated with disappearance of erythromelalgic symptoms (see also section 4.2).[27,28]

Both anagrelide and its active metabolite inhibit platelet cyclic adenosine monophosphate phosphodiesterase (PDE)III and phospholipase A_2,[15,16] an effect that is independent of the effects on megakaryocyte differentiation.[16] 3-Hydroxy anagrelide, is nearly 40 times more potent than anagrelide in inhibiting PDEIII, an effect with possible cardiovascular consequences (see section 2.2).[17] While significant inhibition of platelet aggregation appears only to occur at plasma anagrelide concentrations higher than those required for the thrombocytopenic effect, 3-hydroxy anagrelide has some anti-aggregatory activity at therapeutic concentrations as suggested by its greater anti-PDEIII potency.[17] The effects of anagrelide on the abnormal platelet aggregation seen in some patients with essential thrombocythaemia are as yet unresolved. Spontaneous aggregation in unmodified whole blood and defective platelet aggregation (i.e. no response to epinephrine and/or low response to adenosine diphosphate or collagen) were not improved by anagrelide in several small studies in patients with essential thrombocythaemia.[3,15,24] However, spontaneous platelet aggregation (measured in platelet-rich plasma using an aggregometer) occurring in six of 17 patients with essential thrombocythaemia before treatment with anagrelide did not occur in five of these six after anagrelide-induced normalisation of platelet counts.[29]

While baseline plasma platelet-derived growth factor (PDGF) levels were higher and intraplatelet PDGF levels were lower in 15 patients with essential thrombocythaemia than in healthy controls, these returned to near normal levels in most patients receiving anagrelide for ≥2 months.[30] Anagrelide had little effect on the increased plasma levels of other growth factors involved in myelofibrosis (transforming growth factor-β and basic fibroblast growth factor) in these patients.[30] In line with these results, there was no evidence of a stimulating effect of anagrelide, after a median duration of about 2 years, on the progression of myelofibrosis as assessed using bone marrow biopsies in two small studies (one prospective: n = 17;[31] one retrospective: n = 15[20]).

While there are no reports of increased bleeding associated with anagrelide monotherapy,[21] there are indications that the addition of aspirin to anagrelide treatment can synergistically increase the incidence of bleeding in patients with essential thrombocythaemia (section 4.2.1),[32-35] possibly as a result of the known effects of aspirin being aggravated by the vasodilating effect of anagrelide (section 2.2). In a 6-month study in patients with myeloproliferative disorders (n = 97), bleeding events occurred in nine patients receiving anagrelide plus concomitant low-dose aspirin versus two receiving anagrelide alone (figure 1).[36]

2.2 Cardiovascular Effects

Anagrelide-associated decreased peripheral vascular resistance and blood pressure and increased heart rate and positive inotropic effects (increased

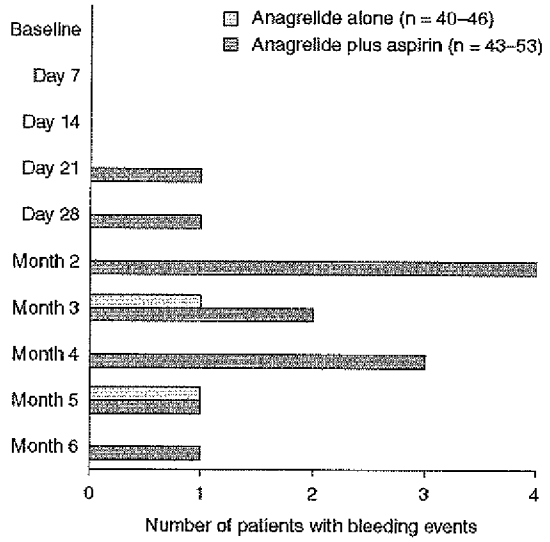

Fig. 1. Bleeding events in patients receiving oral anagrelide with and without concomitant aspirin. Patients with myeloproliferative disorders (n = 97) who were at high risk of complications if untreated or who were refractory to or intolerant of previous treatment with hydroxycarbamide or interferon-α received anagrelide 1–2 mg/day for 6 months, with or without concomitant low-dose aspirin (numbers receiving aspirin varied during treatment), in a noncomparative prospective trial.[36] Some patients had more than one bleeding event.

ventricular contractility) noted in preclinical studies have since been confirmed in humans, although blood pressure appears to return to baseline levels during maintenance therapy.[3,19] Vasodilation, as shown by increases in microvascular area in anagrelide recipients[20] as a consequence of the PDEIII-inhibiting effects of the drug, can cause headache, fluid retention, dizziness and postural hypotension, and occasionally serious cardiovascular disease such as congestive heart failure (section 5).[19,37,38]

The main contributor to the cardiovascular effects of anagrelide is likely to be the active metabolite, 3-hydroxy anagrelide.[17] Because of the inhibitory effect of anagrelide and its metabolite on PDEIII, concomitant administration of drugs such as milrinone, enoximone, amrinone, olprinone or cilostazol may result in exacerbation of their effects.[34]

3. Pharmacokinetic Properties

Most of the pharmacokinetic data on oral anagrelide in this section were obtained from the Citizen Petition submitted to the US FDA by the manufacturers[17] or the manufacturers' prescribing information.[34,35]

3.1 General Properties

An overview of the pharmacokinetic properties of anagrelide is given in table I. The kinetics are linear in the dose range 0.5–2mg.[35] Peak plasma anagrelide concentrations (C_{max}) are reached in 2 hours in patients with essential thrombocythaemia (table I), and anagrelide does not accumulate in plasma with repeated administration.[35] Absorption of anagrelide is extensive, as evidenced by recovery in the urine of >70% of an oral 1mg radiolabeled dose to healthy volunteers.[39] However, the absolute oral bioavailability is thought to be <50%.[17]

The terminal half-life of the parent compound is 1.7 hours.[17] Anagrelide is rapidly metabolised to two main metabolites, the active 3-hydroxy anagrelide (BCH24426) and the inactive 5,6-dichloro-3,4-dihydroquinazol-2-ylamine (RL603) [figure 2].[16,17] 3-Hydroxy anagrelide is largely formed via first-pass metabolism by the cytochrome P450 (CYP)1A2 enzyme.[17] Exposure to 3-hydroxy anagrelide is higher than to the parent compound (table I).[17] In patients with essential thrombocythaemia, systemic exposure was higher and the elimination half-life was longer than in healthy volunteers (table I).

Table I. Overview of anagrelide pharmacokinetics. Mean variables for anagrelide and its active metabolite 3-hydroxy anagrelide were normalised to a 1mg dose and 70kg bodyweight after oral administration to adult healthy volunteers and patients with essential thrombocythaemia (ET)[17]

	Healthy volunteers (n = 38)		Patients with ET (n = 18)	
	anagrelide	3-hydroxy anagrelide	anagrelide	3-hydroxy anagrelide
AUC_∞ (ng • h/mL)	11.1	18.0	19.5	44.1
C_{max} (ng/mL)	5.0	5.5	6.2	8.7
t_{max} (h)	1.3	1.3	2.0	2.3
$t_{½}$ (h)	1.5	2.5	1.7	3.9

AUC_∞ = area under the plasma concentration-time curve from time zero to infinity; C_{max} = maximum plasma concentration; t_{max} = time to C_{max}; $t_{½}$ = elimination half-life.

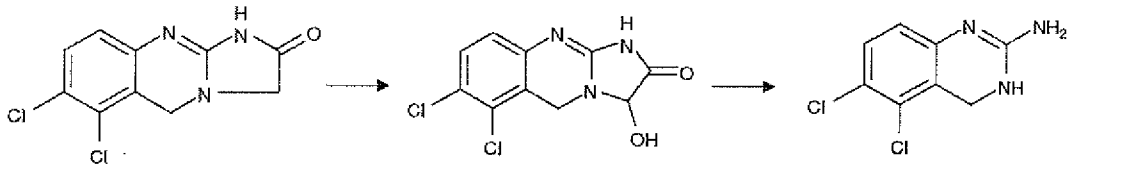

Fig. 2. Summary of the metabolic pathway of anagrelide in healthy volunteers.[16]

The effect of food on the bioavailability of anagrelide is complex. While the C_{max} of the parent compound is decreased by about 14% and the area under the plasma concentration-time curve (AUC) is increased by about 20% in the presence of food, C_{max} of 3-hydroxy anagrelide is decreased by about 30% and AUC is unaffected by food.[17]

Less than 1% of unchanged drug is recovered in the urine.[34] The elimination of anagrelide could be slowed by interaction with drugs such as fluvoxamine, or by grapefruit juice (which inhibit CYP1A2).[34] Similarly, the elimination of drugs like theophylline could be affected by concomitant ingestion of anagrelide.[34] There is no evidence of a pharmacokinetic interaction between anagrelide and hydroxycarbamide, digoxin or warfarin.[34]

3.2 Special Patient Groups

The time to C_{max} and the elimination half-lives of anagrelide, 3-hydroxy anagrelide and 5,6-dichloro-3,4-dihydroquinazol-2-ylamine were similar for paediatric (aged 7–14 years) and adult (aged 16–86 years) patients with thrombocythaemia associated with a myeloproliferative disorder.[35] However, dose- and bodyweight-normalised exposure (C_{max} and AUC) for anagrelide in paediatric patients was about half that in older adult patients, implying more efficient clearance in the younger age group.[35]

While severe renal impairment does not appear to affect the pharmacokinetics of anagrelide, total exposure (AUC) to anagrelide after a single 1mg dose was 8-fold higher in subjects with moderate hepatic impairment than in healthy subjects.[35,40]

4. Therapeutic Efficacy

While response rates involving control of the platelet count alone have been useful for phase II studies of treatment of essential thrombocythaemia, investigations at later stages should assess changes in the natural history of the disease and alleviation of disease-related complications, with associated improvements in quality of life. The main focus of the early noncomparative trials of oral anagrelide in patients with essential thrombocythaemia was on reduction of platelet counts (section 4.1); however, the focus of the one available randomised trial (the Primary Thrombocythaemia 1 [PT1] trial)[33] was on the effects of anagrelide or hydroxycarbamide on the occurrence of vascular events (section 4.2).

In the nonblind, multicentre PT1 trial, high-risk adult patients with essential thrombocythaemia were randomised to treatment with oral anagrelide at an initial dosage of 0.5mg twice daily (n = 405) or hydroxycarbamide 0.5–1 g/day (n = 404) for a median of 39 (range 12–72) months.[33] Dosages of both drugs were adjusted to maintain platelet counts at $<400 \times 10^9$/L. All patients received concomitant aspirin 75–100 mg/day (n = 792) or, if aspirin was contraindicated, dipyridamole (n = 13) or clopidogrel (n = 4) [dosages not reported]. Diagnosis was made using the criteria of the PVSG.[6] The patients were at high risk of thrombotic or haemorrhagic vascular events (criteria included age >60 years, platelet count $\geq 1000 \times 10^9$/L, or a history of ischaemia, thrombosis, embolism or haemorrhage caused by essential thrombocythaemia, or hypertension or diabetes mellitus requiring drug therapy). Patients were excluded from the study if they had the t(9;22) translocation or *BCR-ABL* fusion gene, myelodysplasia, myelofibrosis or causes of thrombocytosis,

Table II. Efficacy of oral anagrelide in patients (pts) with essential thrombocythaemia (ET). Summary of noncomparative, prospective clinical studies in pts (aged 14–83 [median 33–59] years) with myeloproliferative disorders including or restricted to ET

Study	Diagnosis and no. of evaluable pts [% with symptoms]	Previously[a] untreated pts (%)	PC (× 10⁹/L) [mean unless otherwise indicated]	Pts receiving concomitant therapy	Anagrelide dosage (mg/d)[b] initial	maintenance [mean]	Duration of therapy	Response defined as reduction in PC[c] to:	Response rate (%)
Basara et al.[41][d]	ET 20, PV 14, CML 4 [NR]	NR	[1220]	NR	20	NR	NR	<600 or 50%	95
Birgegård et al.[14]	ET 42, PV 17, MF 1 [NR]	55	>600 (SP) or >1000 (AP)	ASP: 47%	1	[2.2]	2y	CR: <400 (SP) or <600 (AP); PR: ≤50%	CR: 67; PR: 6; ETCR: 76; ETPR: 6
Kornblihtt et al.[42]	ET 54 [67]	44	520–2206 [995]	NR	1–3	0.5–5.5 [1.5]	0.8–14.3 [med 6] y	CR: <400; PR: 400–600	CR: 78; PR: 18
Mazzucconi et al.[43]	ET 39 [31]	62	>650 (SP) or ≥900 (AP) [1197]	ASP: 0	1	1–3 [2]	0.5–12.5 [med 6] y	CR: <450; PR: 450–600	CR: 38; PR: 44[e]
Mazur et al.[44]	ET 40 [NR]	0	[1136]	NR	NR	1–3.5 [2]	8–54 [med 23] mo	CR: ≤450	CR: 55; PR: 43
Mills et al.[45]	ET 16 [81]	0	>600 [728]	ASP: 31%; WAR: 38%; HDC: 13%	1–2	1–3 [1.9]	3mo	CR: <400; PR: <600	CR 44; PR 44
Petrides et al.[46]	ET 46 [85]	33	>600 (SP) or >900 (AP) [1193]	ASP: 54%	2	1–12 [3.1]	1–84 [med 12.5] mo	CR: ≤600 and 50%; PR: 50–80%	CR: 88; PR: 6
Steurer et al.[36]	ET 79, PV 16, MF 2 [57[f]]	29[g]	335–1912 [med 743]	ASP: 55%; IFN or HDC: 20%	1	0.5–4.5 [med 2]	6mo	CR: <450; VGPR: <600; PR: <50% but >600	CR: 52; VGPR: 26; PR: 2

a In most trials, patients were transferred from previous therapy because of lack of efficacy or occurrence of unacceptable adverse events.[36,43-46]

b Given in divided doses.

c Reductions are given as goal PC x 10⁹/L or as a percentage of baseline PC. Where specified, the reduction was to be maintained for ≥4wk.[14,36,41,43,46]

d Abstract.

e Of nine pts with high baseline PC (≥1500 x 10⁹/L), six achieved CR and two PR.

f Of these, 29 patients (30%) had thromboembolic complications (6% major and 24% minor) during the 6mo before the study.

g Newly diagnosed pts had to be high-risk for inclusion.

AP = asymptomatic pts; **ASP** = aspirin; **CML** = chronic myeloid leukaemia; **CR** = complete response; **ETCR/ETPR** = CR or PR in pts with ET; **HDC** = hydroxycarbamide; **IFN** = interferon-α; **med** = median; **MF** = myelofibrosis; **NR** = not reported; **PC** = platelet count; **PR** = partial response; **PV** = polycythaemia vera; **SP** = symptomatic pts; **VGPR** = very good partial response; **WAR** = warfarin.

breathlessness, cardiac pain, myocardial infarction in the previous 3 months, severe congestive heart failure, severe ventricular arrhythmia or leg ulceration, or were pregnant/lactating. About 30% had received previous hydroxycarbamide treatment and 18% had received no prior treatment.[33]

The composite primary efficacy endpoint in this study comprised death from thrombosis or haemorrhage, or occurrence of arterial thrombosis, venous thrombosis or serious haemorrhage.[33] Secondary endpoints included occurrence of the first arterial or venous thrombotic event, serious haemorrhage or death, incidence of transformation to myelofibrosis, acute myeloid leukaemia, myelodysplasia or polycythaemia vera, and platelet count. All endpoints were independently reviewed/validated by clinicians blinded to treatment assignment.

Of the reviewed noncomparative trials, eight were prospectively designed (table II) and two were multicentre;[14,36] patients were followed for up to 12.5 years. Three of these trials enrolled patients with other myeloproliferative disorders (i.e. polycythaemia vera, chronic myeloid leukaemia, myelofibrosis); however, in these trials, most patients (53–81%; n = 20–79) had essential thrombocythaemia.[14,36,41] The other trials limited enrolment to patients with essential thrombocythaemia (n = 16–48).[42-46] Diagnosis was mostly made using the criteria of the PVSG,[6] although one trial[14] used the criteria of Kutti and Wadenvik which include bone marrow histology.[47] In selected trials, entry was restricted to patients who had been previously treated with myelosuppressive agents,[45] or patients with essential thrombocythaemia refractory to previous hydroxycarbamide.[44] The proportion of previously untreated patients ranged from 29% to 62% in the other trials, where reported.[14,36,42,43,46] Two trials reported inclusion of a washout period before initiation of anagrelide therapy.[14,43] Anagrelide was given initially at 1–2 mg/day (most studies indicated divided doses) and maintenance doses were adjusted thereafter to achieve target platelet counts (table II). The primary endpoint (reduced platelet counts) was defined in two trials.[36,43]

One retrospective study has been included because of its size.[2] Patients in this study had thrombocythaemia associated with myeloproliferative disorders, were aged 5–98 years and received an initial anagrelide dosage of 2 mg/day. The 934 patients with essential thrombocythaemia in this trial were followed for >7 years; 18% of these patients received concomitant hydroxycarbamide and many received aspirin during the study; 76% had received previous medication; 31% were receiving anagrelide because of poor platelet control with previous therapy.[2]

Although patient numbers were small (n = 35), a second retrospective study was included because 27 of the 33 responding patients had received anagrelide for an extended period (7–16 [median 11] years).[48] Younger patients (aged 17–48 years) were chosen for this trial to circumvent confounding problems associated with age and comorbid conditions. Before treatment, 27 patients had symptomatic disease (history of thrombosis 20%, haemorrhage 26%, vasomotor manifestations 51%), and 24 patients had received hydroxycarbamide or busulfan therapy. Oral anagrelide was administered at an initial dosage of 1–10 mg/day.

4.1 Platelet Counts

4.1.1 Prospective Noncomparative Trials

Anagrelide reduces platelet counts in patients with essential thrombocythaemia. Although response criteria in all the trials required reduced platelet counts, the exact definitions of complete and partial response varied among the trials, and results specific to patients with essential thrombocythaemia were not given in all trials (see table II). Nonetheless, the overall (complete plus partial) response rates tended to be high for the reviewed trials, ranging from 82% to 98% in patients with essential thrombocythaemia.[14,42-46] Complete responses were seen in 38–88% of patients with essential thrombocythaemia.[14,42-46] In two small trials of patients with essential thrombocythaemia refractory to previous treatment (mainly hydroxycarbamide), complete response rates were 44% and 55%, and overall response rates were 88% and 98%.[44,45]

The time to reach a response, where reported, varied from 6 days to 12 months (median, given in four trials, ranged from 2 weeks to 4 months[14,42-44]);[14,41,43-45] this broad range was possibly dependent to some extent on variations in the rate of dosage increase during the early stages of therapy. Some response is usually reached within the first 2 weeks.[42]

4.1.2 Retrospective Studies

The large retrospective study showed that a clinically significant reduction (complete plus partial response) in platelet counts occurred in 79% of patients with essential thrombocythaemia on receipt of oral anagrelide therapy (83% of previously untreated patients).[2] A complete response (the primary endpoint; 67% of patients) was defined as reduction in platelet count of ≥50% from baseline or to

$\leq 600 \times 10^9$/L for ≥4 weeks. A partial response (12%) was defined as reductions of 20–50% from baseline. Mean platelet counts were reduced from 1090×10^9/L to $<600 \times 10^9$/L at 2 months and to $470–480 \times 10^9$/L from the end of the first year.[2]

The initial overall response to anagrelide in the long-term retrospective study in younger patients[48] was 94%, comprising complete remission (a sustained platelet count of $<450 \times 10^9$/L) in 26 patients, partial remission ($450–600 \times 10^9$/L) in seven patients and no response in two patients.[48] Of the 29 evaluable patients who received anagrelide (at a maintenance dosage of 1–5 [median 2.5] mg/day) for the duration of the study, 19 (66%) were in complete remission and ten (34%; six of these had initially experienced complete remission) were in partial remission at final follow-up.

4.1.3 Versus Hydroxycarbamide

Anagrelide and hydroxycarbamide, given with concomitant aspirin to high-risk patients, reduced platelet counts to a similar extent at 9 months of treatment and thereafter (from baseline medians of 930 and 947×10^9/L, respectively, to 370×10^9/L for both; estimated from a graph).[33] However, at the earlier timepoints of 3 and 6 months, platelet counts were lower in the group receiving hydroxycarbamide plus aspirin (370 and 330×10^9/L) than in those receiving anagrelide plus aspirin (450 and 370×10^9/L) [estimated from a graph; $p < 0.001$ for both 3 and 6 months]. The numbers of patients achieving platelet counts $<400 \times 10^9$/L at various timepoints was not reported.

4.2 Thrombohaemorrhagic Symptoms/Events

4.2.1 Versus Hydroxycarbamide

Arterial or venous thrombosis, serious haemorrhage or death from vascular causes (composite primary endpoint) occurred more often in recipients of anagrelide plus aspirin than in those receiving hydroxycarbamide plus aspirin (55 vs 36 patients; odds ratio 1.57; 95% CI 1.04, 2.37; $p < 0.05$; figure 3) in the PT1 trial.[33] The estimated risk (after a median of 39 months' follow-up) of this endpoint occurring at 5 years of treatment was 16% with anagrelide versus 11% with hydroxycarbamide.

The groups were matched for baseline thrombohaemorrhagic experience and time from diagnosis to enrolment; 6% had previous venous thromboembolism, 18% had previous arterial thrombosis and 6% had previous haemorrhage. The overall secondary endpoint of venous thrombosis was more common in hydroxycarbamide plus aspirin recipients than in those receiving anagrelide plus aspirin (14 [4%] vs 3 [1%]; $p < 0.01$), while arterial thrombosis (17 [4%] vs 37 [9%]) and serious haemorrhage (8 [2%] vs 22 [5%]) each occurred significantly more often in recipients of anagrelide plus aspirin (both $p < 0.01$). However, the study was not powered to differentiate to this level, and these results may have been subject to selection bias or chance.

Among the individual parameters making up the secondary endpoints (figure 3), a significant difference in favour of anagrelide plus aspirin was seen for deep-vein thrombosis (1 vs 9 patients; $p < 0.01$). Significant differences in favour of hydroxycarbamide plus aspirin were seen for transient ischaemic attack (1 vs 14 patients; $p < 0.001$) and gastrointestinal bleeding (3 vs 13 patients; $p = 0.01$). There were no significant differences between the groups for other arterial thromboses (myocardial infarction, stroke or unstable angina), venous thromboembolisms (pulmonary embolism or hepatic-vein thrombosis) or serious haemorrhages (nasal, intracranial or other), and there were no significant differences between the groups in the incidence of death from thrombosis or haemorrhage.

4.2.2 Noncomparative Trials

Five of the prospective trials included an indication of the number of patients (31–85% of the total cohorts; table II) with thrombohaemorrhagic symptoms often associated with thrombocythaemia (e.g. microvascular circulation disturbances, transient ischaemic attacks, headache/dizziness, cutaneous symptoms, etc.) at baseline or a history of thrombohaemorrhagic events (e.g. stroke, myocardial or pulmonary infarction, bleeding requiring transfusion).[36,42,43,45,46] Symptoms improved and/or there

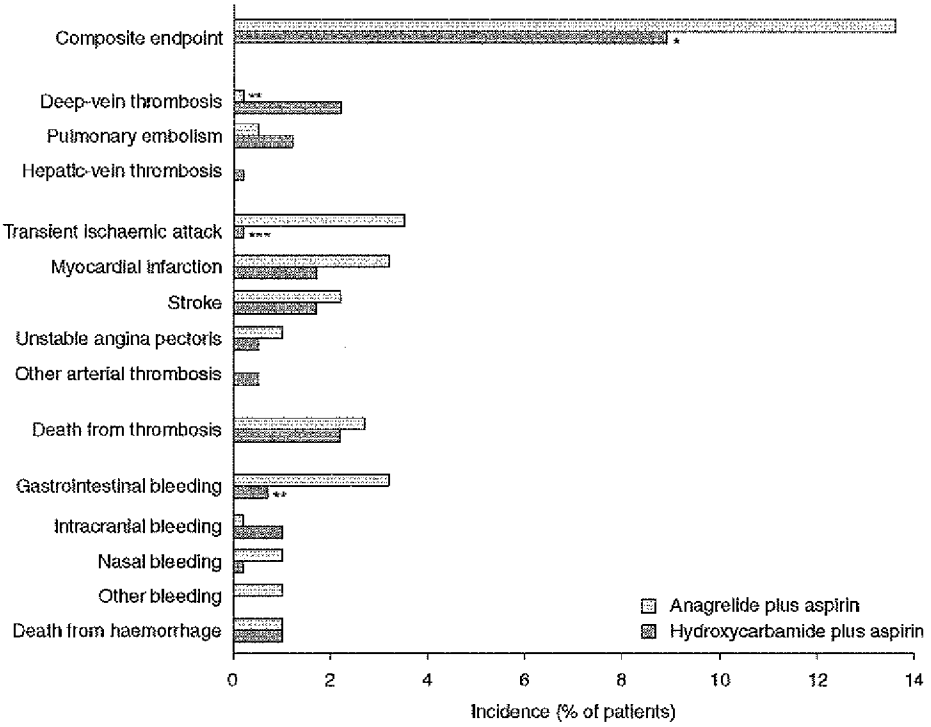

Fig. 3. Comparative efficacy of oral anagrelide plus aspirin in high-risk patients with essential thrombocythaemia. Incidence of venous or arterial thrombosis, serious haemorrhage or death from thrombosis or haemorrhage (the composite endpoint) and individual secondary thrombohaemorrhagic endpoints in patients at high risk of vascular events randomised to treatment with anagrelide (initial dosage 0.5mg twice daily; n = 405) or hydroxycarbamide (initial dosage 0.5–1 g/day; n = 404) for a median of 39 months in a nonblind, multicentre study.[33] Dosages were adjusted to achieve a platelet count of <400 × 10^9/L. All patients received concomitant aspirin, dipyridamole or clopidogrel. * $p < 0.05$, ** $p \leq 0.01$, *** $p < 0.001$ vs comparator.

was no recurrence of previous complications in 50–100% of patients during anagrelide therapy of 3 months to 6 years' duration in the three studies providing this information.[29,36,45] In one study, symptoms disappeared in all patients with a platelet count response during long-term therapy (median 73 months; number of responding patients not reported).[43] Others found that symptoms reappeared in some patients (8–13%) despite continued platelet count reduction.[42,44]

Thrombotic events (none fatal) occurred in seven patients (20%) and major haemorrhagic events (one fatal; three gastrointestinal) occurred in seven patients (20%) during anagrelide treatment in the long term, retrospective study in young patients.[48] In all the affected patients, the platelet count was >400 × 10^9/L at the time of the event.

4.3 Disease Transformation

The rate of development of myelofibrosis was significantly lower in recipients of hydroxycarbamide plus aspirin versus anagrelide plus aspirin in the PT1 trial (figure 4).[33] Comparative baseline histology-based risk factors for transformation to myelofibrosis were not, however, presented. Myelofibrotic transformation was defined as at least grade 3 reticulin fibrosis in a bone marrow biopsy (plus an increase by ≥1 grade from baseline), plus at least two of: increased spleen size by ≥3cm; decreased haemoglobin by ≥20 g/L; immature myeloid or erythroid cells in blood smear; tear-drop poikilocytes in blood smear; or night sweats, bone pain or weight loss of >10% in 6 months.

There were no significant differences between the groups in the rate of transformation to acute

myeloid leukaemia/myelodysplasia or polycythaemia vera or death from transformation (figure 4).[33] However, 39 months' follow-up is possibly inadequate for estimating the total risk in this respect.

Of the 2251 anagrelide recipients with essential thrombocythaemia evaluable for disease transformation analysis in a large retrospective study, 47 patients (2%) developed acute leukaemia/myelodysplastic syndrome, mostly within the first year of treatment.[2] All 47 patients had received previous cytotoxic drug treatment.

4.4 Pharmacoeconomic Considerations

Two pharmacoeconomic direct-cost analyses using Markov models based on noncomparative phase II clinical trial data indicate that, despite its relatively high purchase price, anagrelide could be cost effective in some scenarios.[49,50]

In one study, the cost effectiveness of anagrelide treatment for essential thrombocythaemia improved over 1 year from the first 3 months of treatment ($US2462 per major complication [gastrointestinal bleed, transient ischaemic attack, stroke, preinfarction angina, myocardial infarction] prevented; year of costing not reported) to 10–12 months ($US1505).[49]

The other analysis, conducted from a societal perspective, found that lifelong treatment with anagrelide for a 40-year-old man with essential thrombocythaemia was associated with a marginal cost effectiveness versus hydroxycarbamide of $US71 737 per year of life gained.[50] If hydroxycarbamide was associated with an assumed risk of disease transformation to leukaemia of 0.1 and the threshold willingness for society to pay was $US75 000, anagrelide was optimal in 66% of 1000 trials; however, hydroxycarbamide was optimal in all trials when the willingness to pay was reduced to $US50 000. Further, if the hydroxycarbamide-associated risk of transformation to leukaemia was reduced to 0.05, the incremental cost effectiveness of anagrelide increased to $US156 969 per additional life-year gained. Interferon-α was associated with higher costs and lower efficacy than anagrelide. The study used an annual discount rate of 3% and was published in 2002 (year of costing not reported).

5. Tolerability

Information for this section was drawn mainly from the manufacturers' prescribing information[34]

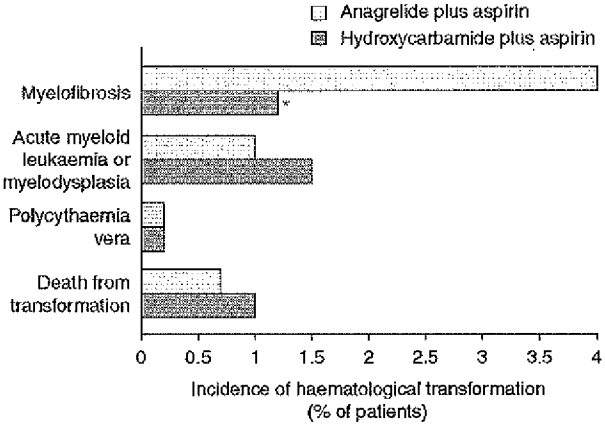

Fig. 4. Comparative haematological transformation with anagrelide plus aspirin in patients with essential thrombocythaemia at high risk of vascular events. Patients were randomised to treatment with oral anagrelide (initial dosage 0.5mg twice daily; n = 405) or hydroxycarbamide (initial dosage 0.5–1 g/day; n = 404) for a median of 39 months in a nonblind, multicentre study.[33] Dosages were adjusted to achieve a platelet count of <400 × 10^9/L. All patients received concomitant aspirin, dipyridamole or clopidogrel. Transformation occurred in the anagrelide and hydroxycarbamide groups at a median of 45 and 34 months after diagnosis for myelofibrosis and 83 and 36 months after diagnosis for acute myeloid leukaemia. * $p < 0.05$.

and selected clinical studies[2,14,33,48] (see section 4 for design details).

5.1 General Profile

The tolerability profile of anagrelide at a mean dosage of about 2 mg/day for up to 4 years in 942 patients from three noncomparative studies and 3660 patients in a further noncomparative study (34 took anagrelide for up to 5 years) has been outlined by the manufacturers in the prescribing information.[34,35] The most common adverse events in the 942-patient cohort were headache (44%), palpitations (26%), diarrhoea (26%), asthenia (23%), oedema (21%), nausea (17%), abdominal pain (16%) and dizziness (15%).[35] Other common events occurring in anagrelide recipients at an incidence of 5–15% included other pain, dyspnoea, flatulence, vomiting, fever, peripheral oedema, rash, anorexia, tachycardia, pharyngitis, malaise, cough, paraesthesia, pruritus and dyspepsia; most adverse events were mild in intensity.[35] Serious adverse events that occurred rarely during anagrelide therapy included congestive heart failure, myocardial infarction, cardiomyopathy, cardiomegaly, complete heart block, atrial fibrillation, cerebrovascular accident, pericarditis, pericardial effusion, pleural effusion, pulmonary infiltrates, pulmonary fibrosis, pulmonary hypertension, pancreatitis, gastric/duodenal ulceration and seizure.[35] The incidence/severity of adverse events in anagrelide recipients is dose-dependent.[14] The incidence of serious adverse events was twice as high in patients aged ≥60 years than in younger patients in a retrospective study of 2251 patients with essential thrombocythaemia.[2]

Anaemia occurs commonly in anagrelide recipients,[34] possibly as a result of the vasodilating effects of anagrelide.[14] Mean haemoglobin levels dropped significantly and dose-dependently in the first few weeks of anagrelide treatment (from a baseline of 13.2 g/dL to a nadir of 12.7 g/dL in week 8), and remained lower than baseline during a prospective, 2-year trial in 60 patients with myeloproliferative disorders (70% with histologically confirmed essential thrombocythaemia).[14] In contrast, anaemia developed later, after approximately 1–10 years of anagrelide treatment, in four of 39 patients (aged 22–52 years) with histologically confirmed essential thrombocythaemia in another prospective trial.[43] Thrombocytopenia and pancytopenia are listed by the manufacturers as uncommon (<1% of patients).[34]

The incidence of adverse events falls with extended treatment in some patients. For example, in a retrospective study of younger patients (aged 17–48 years) who received anagrelide for up to 16 (median 11) years, the incidence of headache dropped from 34% in the first 3 months to 6% long term.[48] Similarly, initial and long-term incidences of tachycardia were 23% and 9%, of oedema were 14% and 6% and of diarrhoea were 9% and 0%. However, a prospective, noncomparative, 2-year study (n = 60),[14] with tolerability as one of the primary objectives, found that although adverse events abated to some extent during the initial months of therapy, the intensity of adverse events remained unchanged in many patients with prolonged treatment. In fact, withdrawal from anagrelide treatment because of adverse events was high in this study: patients discontinued treatment because of insufficient efficacy at a tolerable dosage (n = 13), adverse events despite complete response (n = 10) or other reasons unrelated to tolerability (n = 7). Fifty percent of patients remained on anagrelide treatment after 2 years. The most common and severe adverse events in this study were headache, palpitations and diarrhoea.

The number of patients withdrawing from treatment because of adverse events in the randomised PT1 trial in 809 high-risk patients with essential thrombocythaemia was higher in those receiving anagrelide plus aspirin than in those receiving hydroxycarbamide plus aspirin (22% vs 11%; $p < 0.001$).[33] Headache (13% vs 2%; $p < 0.001$), diarrhoea (4% vs 1%; $p = 0.01$), abdominal pain (2% vs <1%; $p < 0.01$) and noncardiac oedema (6% vs 1%; $p < 0.001$) were more common with anagrelide than with hydroxycarbamide. However, dermatological events such as leg or mouth ulcers (11% vs 7%; $p = 0.05$) and diabetes (2% vs 1%; $p = 0.05$) occurred more often with hydroxycarbamide than with anagrelide. From 3 months after initiating treatment,

median white-cell counts were significantly and persistently lower in hydroxycarbamide recipients than in anagrelide recipients. There were no significant differences between the groups in the incidence of minor haemorrhage, nausea/vomiting, peptic ulcer/oesophagitis/gastritis, anaemia or thrombocytopenia/neutropenia.

5.2 Cardiovascular Effects

While palpitations, tachycardia and fluid retention were common in patients receiving anagrelide in studies monitored by the manufacturers (occurring in 1–10% of patients), congestive heart failure, hypertension, arrhythmia, atrial fibrillation, supraventricular/ventricular tachycardia, syncope, oedema and chest pain were uncommon (0.1–1%), and angina pectoris, myocardial infarction, cardiomegaly, pericardial effusion, vasodilatation, migraine and postural hypotension were rare (0.01–0.1%) [see also section 2.2].[34]

Palpitations (including irregular pulse) occurred in 16% and 2% of anagrelide and hydroxycarbamide recipients in the PT1 trial ($p < 0.001$).[33] There were no significant differences between the groups in the incidence of cardiac failure or arrhythmia.

6. Dosage and Administration

Anagrelide is available in Europe, the US and numerous other countries worldwide. In Europe, anagrelide has orphan drug status and is indicated for the reduction of elevated platelet counts in at-risk patients with essential thrombocythaemia who are intolerant of or refractory to their current therapy. Patients at risk are defined as those >60 years of age, those with a platelet count $>1000 \times 10^9$/L, or those with a history of thrombohaemorrhagic events.[34] In the US, anagrelide is indicated for the treatment of patients with thrombocythaemia associated with myeloproliferative disorders, to reduce the elevated platelet count and the risk of thrombosis and to ameliorate associated symptoms including thrombohaemorrhagic events.[35]

The anagrelide package inserts currently recommend a starting oral dosage for adults of 0.5mg twice daily (Europe) or 0.5mg four times daily or 1mg twice daily (US); this dosage should be maintained for ≥1 week and thereafter titrated individually to achieve a platelet count $<600 \times 10^9$/L and ideally $150–400 \times 10^9$/L.[34,35] Although experience in children is limited, starting dosages have ranged from 0.5 mg/day (recommended in the US[35]) to 0.5mg four times daily. Dosages should be increased by no more than 0.5 mg/day in a single week, and should not exceed 10 mg/day. The recommended maximum single dose is 2.5mg.

Patients with cardiovascular disease (see also section 2.2) or hepatic dysfunction (section 3.2) should be monitored closely during anagrelide therapy. Dosage reduction is recommended for patients with moderate hepatic impairment in the US and the drug is contraindicated in patients with moderate or severe hepatic impairment in Europe. There is no contraindication or warning in the US for the use of anagrelide in patients with renal impairment but the drug is contraindicated in both moderate and severe impairment (creatinine clearance <50 mL/min) in Europe.[34,35,40]

In addition to the potential pharmacokinetic interactions with drugs inhibiting CYP1A2 outlined earlier (section 3.1), anagrelide may also have synergistic anti-aggregatory effects when given with aspirin (section 2.1). For additional dosage and administration information, the local manufacturer's prescribing information should be consulted.

7. Place of Anagrelide in the Management of Essential Thrombocythaemia

The primary aim of treatment for essential thrombocythaemia is to reduce the incidence of the main causes of morbidity and mortality: thrombohaemorrhagic events and disease transformation to myelofibrosis or acute leukaemia.[13] Because of the adverse effects associated with cytoreductive agents, particularly the leukaemogenic propensity of some, the treatment of low-risk patients remains controversial, with many advocating administration of cytotoxic therapy on the basis of their risk for developing vascular events (i.e. no treatment for low-risk patients).[12]

Table III. Recommendations from Italian haematology societies[13] on specific patient groups requiring platelet-lowering therapy for essential thrombocythaemia

All patients aged >60 years
All patients aged <60 years with a history of major thrombotic or haemorrhagic events
All patients with platelet counts >1500 × 10^9/L
Patients aged 40–60 years with platelet counts >1000 × 10^9/L and a cardiovascular risk factor or familial thrombophilia
Patients aged <40 years with prothrombotic comorbidity (homocysteinuria, familial dominant hypercholesterolaemia)
Patients with severe microcirculatory symptoms

The available treatment guidelines outline the at-risk patient groups for whom platelet-lowering therapy is recommended in Italy (table III).[13] Consensus could not be reached on whether patients aged 40–60 years who have platelet counts <1000 × 10^9/L and no history of major thrombohaemorrhagic events but who have a cardiovascular risk factor or familial thrombophilia should be treated. The associated guidelines for choice of platelet-lowering agent in specific treatment-eligible patient groups, outlined in table IV, were prepared by an Italian expert panel and advisory committee[13] at a time when reliable comparative scientific evidence was scarce. Guidelines for the treatment of essential thrombocythaemia remain in a state of flux as data from ongoing long-term trials become available and diagnostic and classification changes are assessed.

Treatment of essential thrombocythaemia traditionally involves administration of platelet-lowering agents and/or anti-platelet agents (e.g. aspirin, clopidogrel). Anti-platelet agents have been recommended for patients with microcirculatory symptoms if there are no contraindications and for patients with recent major arterial vascular events or coronary artery disease if no previous significant bleeding has occurred.[13]

The most commonly used platelet-lowering agents other than anagrelide are hydroxycarbamide, a nonspecific myelosuppressive agent that reduces all myelogenous lineages via inhibition of DNA synthesis, and recombinant interferon-α, a cytokine that inhibits the growth of multipotent haematopoietic progenitor cells and megakaryocyte-forming units.[13] Hydroxycarbamide reduced the incidence of thrombotic episodes versus no treatment in pa-

Table IV. Italian guidelines for choice of platelet-lowering agent in patients with essential thrombocythaemia who are eligible for such treatment[13]

Patient group	First-line therapy	Alternative therapy[a]	Other factors
Patients aged <40y, no child-bearing potential	IFNα or ANA	HDC	HDC not recommended in very young patients
Patients aged 40–60y, no child-bearing potential, with a history of a major thrombotic event	HDC		
Patients aged 40–60y, no child-bearing potential, without a history of a major thrombotic event	IFNα or ANA[b]		Use of ANA should be monitored by entry into trials or a register
Women with child-bearing potential	IFNα	ANA (second-line) → HDC (third-line)	Patients should stop taking ANA or HDC if pregnancy is suspected
During pregnancy[c]	IFNα		
Patients starting therapy aged 60–70y	HDC	BUS or PIP	Patients already successfully receiving IFNα or ANA should continue
Patients aged >70y	HDC, BUS or PIP		Patients already successfully receiving IFNα or ANA should continue

a Second-line or subsequent treatment options if adverse effects affect quality of life or high doses increase toxicity.

b Since publication of the PT1 trial,[33] some members of the Italian group are now considering HDC plus aspirin as first-line therapy in this patient group.[12,32]

c Treatment is recommended if there is a history of major thrombosis or haemorrhage, platelet counts >1000 × 10^9/L, familial thrombophilia or cardiovascular risk factors.

ANA = anagrelide; **BUS** = busulfan; **HDC** = hydroxycarbamide; **IFNα** = interferon-α; **PIP** = pipobroman; **PT1** = Primary Thrombocythaemia 1.

tients with essential thrombocythaemia,[51] but is associated with dose-limiting haematopoietic impairment, oral or leg ulcers and other skin lesions.[52] Adverse oral or skin effects, including leg ulcers, occurred at a rate of 26% in 133 patients receiving hydroxycarbamide for >2 years, generally appearing ≥5 years after starting treatment and often necessitating withdrawal from treatment.[53] There remains some uncertainty about whether hydroxycarbamide is leukaemogenic. There were no cases of leukaemic or neoplastic transformation in one study of previously untreated patients with essential thrombocythaemia (aged <50 years) receiving hydroxycarbamide alone for a median of 8 (range 5–14) years.[54] However, other studies report therapy-related leukaemic transformation associated with hydroxycarbamide in previously untreated patients,[55,56] and long-term follow-up (6 years) of patients with essential thrombocythaemia in a randomised study of hydroxycarbamide versus no therapy found that the absent or very low risk of leukaemic progression in untreated patients (no cases in this study) was increased slightly with hydroxycarbamide alone (4%).[57] Further, recent histological evidence supports the potential of this drug for leukaemogenic conversion (section 2.1). The rate of transformation is increased when hydroxycarbamide is given with other agents.[52] Until this issue is clarified, it has been suggested that hydroxycarbamide should not be administered to very young patients (table IV).[13,58]

While interferon-α is not leukaemogenic, it does have to be given parenterally and is associated with severe adverse effects (e.g. fatigue, depression, influenza-like symptoms, elevated liver enzymes, anorexia, alopecia, neuropsychiatric symptoms) often necessitating withdrawal.[52] Interferon-α is recommended by some for cytoreductive treatment of high-risk women planning pregnancy (table IV).

Pipobroman (which has a structural resemblance to alkylating agents) is clinically active in patients with essential thrombocythaemia.[59] However, this and the alkylating agent busulfan have been associated with development of secondary acute leukaemia, myelodysplastic syndromes or solid tumours,[57,59,60] especially in patients receiving concomitant or subsequent hydroxycarbamide.[57] Marketing of pipobroman has been discontinued in the US,[61] and busulfan is now rarely used to treat essential thrombocythaemia.[14]

Anagrelide is clearly an effective platelet-lowering agent in both untreated patients and those refractory to previous hydroxycarbamide (section 4.1). In addition, this drug tends to normalise platelet function defects associated with essential thrombocythaemia and has anti-angiogenic properties (section 2.1).

The comparative position of anagrelide in the management of essential thrombocythaemia has not been widely studied to date. Interpretation of the results of the recently published PT1 trial of anagrelide plus aspirin versus hydroxycarbamide plus aspirin in high-risk patients is partly hindered by the use of PVSG diagnostic criteria in the study and by its early termination.[33] As outlined earlier (section 1), the use of bone marrow biopsy in the diagnostic process, which was not included in the design of this trial, allows specific subtypes of patients, with differing prognostic outcomes, to be distinguished. Recent studies have found that, of cohorts of patients initially diagnosed according to PVSG criteria, only about one-third had true essential thrombocythaemia according to WHO (histology-based) criteria.[8,9] The remaining patients had thrombocythaemia associated with prefibrotic or early fibrotic chronic idiopathic myelofibrosis. This patient distribution may well have been repeated in the PT1 trial of anagrelide versus hydroxycarbamide.[33] Further, a recent publication indicates that essential thrombocythaemia patients with the Jak2V617F mutation are more likely to benefit from hydroxycarbamide than from anagrelide treatment, a hypothesis that deserves further investigation.[62] While anagrelide does not affect progression of early fibrotic stages to classical myelofibrosis (section 2.1), hydroxycarbamide may slow fibrotic progression, as demonstrated in the PT1 trial (section 4.3). It thus seems reasonable to assume that the reported risk of myelofibrotic transformation in the anagrelide arm

was not related to treatment, but to the heterogeneity of disease in the included patients.

The safety of concomitant platelet-lowering/antiplatelet therapy remains under discussion. The results of the PT1 trial[33] suggest that an interaction between anagrelide and aspirin may have increased the risk of haemorrhage (see also section 2.1), an outcome that requires further research and potential modification of current recommendations. While bleeding has not traditionally been a problem in patients receiving anagrelide monotherapy (section 2.1), gastrointestinal haemorrhage is known to be associated with aspirin use in patients with polycythaemia vera or essential thrombocythaemia.[63] Aspirin may also have exacerbated bleeding in patients with acquired von Willebrand syndrome, common in patients with elevated platelet numbers.[21,63]

The event rates were low in the PT1 trial despite the size of the study cohort and follow-up of over 3 years. Both anagrelide and hydroxycarbamide are effective agents in this respect; both decreased the number of disease-related events compared with historical records for untreated controls.[51] Anagrelide plus aspirin was associated with a decreased rate of deep-vein thrombosis and an increased rate of transient ischaemic attacks versus hydroxycarbamide plus aspirin in high-risk patients with essential thrombocythaemia in the PT1 trial (section 4.2). Since low-dose aspirin is highly effective in the prevention of platelet-mediated microvascular disturbances such as erythromelalgia and transient ischaemic attacks,[4] the excess incidence of transient ischaemic attacks in anagrelide recipients in this trial may have been associated with the known cardiac effects of anagrelide (section 2.2).

Anagrelide is not mutagenic (section 2.1) and there is no evidence to date to suggest that it is leukaemogenic. Further data from well designed trials (using specific diagnostic criteria) on this and thrombohaemorrhagic outcomes would be helpful. A phase III, randomised, single-blind, multicentre comparison of the efficacy and tolerability of anagrelide versus hydroxycarbamide in high-risk previously untreated patients with essential thrombocythaemia (the AOP 03-007 ANAHYDRET [ANAgrelide HYDRoxyurea in patients with Essential Thrombocythaemia] study) is currently under way.[64]

Anagrelide is associated with headache, palpitations and diarrhoea, as well as the development of cardiovascular effects (section 5), necessitating ongoing monitoring of patients and possible dosage adjustments. It has been suggested that the risk of thrombosis is increased in patients who only partially respond to anagrelide (i.e. platelet counts $>400 \times 10^9$/L);[48] there is therefore a need to clarify the optimal target platelet count for patients with essential thrombocythaemia in randomised trials. Since adverse events associated with anagrelide are dose-related, the administration of high enough doses to achieve this target may not be possible in some, particularly elderly, patients.

Because the treatment of essential thrombocythaemia will be long term, and patients with this disorder are expected to achieve near normal lifetimes, long-term efficacy and tolerability of potential medications are important factors for consideration. Experience with anagrelide in children is limited to case reports that indicate potential for efficacy with only minor adverse effects.[65-69] There is some evidence of long-term (11 years) efficacy (section 4.1.2), and reduction in the incidence of adverse effects with time (section 5), in young adults receiving anagrelide.

While two related pharmacoeconomic analyses have indicated that anagrelide is cost effective in some scenarios (section 4.4), these analyses were based on noncomparative clinical trials and were sensitive to the leukaemogenic potential of hydroxycarbamide. Further pharmacoeconomic analyses based on well designed comparative trials are awaited with interest.

In conclusion, anagrelide is well established as an effective platelet-lowering agent in most patients with essential thrombocythaemia, including both treatment-naive patients and those refractory to other cytoreductive therapy. Results of the only randomised trial to date (the PT1 study) indicated that the composite primary endpoint (arterial or ve-

nous thrombosis, serious haemorrhage or death from vascular causes) occurred more often in recipients of anagrelide plus aspirin than in those receiving hydroxycarbamide plus aspirin. This trial also indicated that the incidence of the secondary endpoints transient ischaemic attack and gastrointestinal bleeding favoured hydroxycarbamide plus aspirin, while the incidence of venous thrombosis favoured anagrelide plus aspirin. There were no differences between the groups in the incidence of other arterial or thrombotic embolisms or haemorrhagic secondary endpoints. However, the design of the PT1 study has been queried with respect to the heterogeneous nature of the study population (possible inclusion of patients with early myelofibrotic disease) and the concomitant use of aspirin (interaction with anagrelide causing increased bleeding events). Further data are therefore required before the role of anagrelide in essential thrombocythaemia can be finalised. In the meantime, when considering treatment options for patients with this disorder, anagrelide's positive effects on platelet function, lack of mutagenicity and lack of association with leukaemia or angiogenesis must be balanced against its comparative expense and positive inotropic effects. Thus, the role of anagrelide in the management of high-risk patients with essential thrombocythaemia will ultimately depend on individual patient assessment and future clarification of the potential leukaemogenicity of hydroxycarbamide.

Disclosure

During the peer review process, the manufacturer of the agent under review was also offered an opportunity to comment on this article; changes based on any comments received were made on the basis of scientific and editorial merit.

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EXHIBIT 3

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Comparison of the biological activities of anagrelide and its major metabolites in haematopoietic cell cultures

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1 The platelet-lowering drug anagrelide inhibits bone marrow megakaryocytopoiesis by an unknown mechanism. Recently, it was found that anagrelide is bio-transformed in humans into two major metabolites (6,7-dichloro-3-hydroxy-1,5 dihydro-imidazo[2,1-b]quinazolin-2-one (BCH24426) and 2-amino-5,6-dichloro-3,4,-dihydroquinazoline (RL603). Whether these metabolites have biological activities that may underlie the mode of action of the parent drug is presently unclear. To clarify this question here we have compared the activities of anagrelide, BCH24426 and RL603 on the growth and differentiation of $CD34^+$ haematopoietic progenitor cells in liquid culture and on the migration of differentiated megakaryocytes.

2 Incubation with either anagrelide, BCH24426 or RL603 did not affect the early expansion of $CD34^+$ cells stimulated by thrombopoietin. In contrast, both anagrelide and BCH24426 potently inhibited the development of megakaryocytes ($IC_{50} \pm$ s.e.m. $= 26 \pm 4$ and 44 ± 6 nM, respectively), whereas RL603 showed no significant effect.

3 Anagrelide and BCH24426 did not affect erythroid or myelomonocytic differentiation stimulated by erythropoietin or granulocyte–macrophage colony-stimulating factor, demonstrating the selectivity of these compounds against the megakaryocytic lineage.

4 Neither anagrelide nor its metabolites showed a significant effect on the migratory response of megakaryocytes towards stromal cell-derived factor-1α.

5 Although BCH24426 was shown to be considerably more potent than anagrelide as an inhibitor of phosphodiesterase type III (PDEIII) ($IC_{50} = 0.9$ vs 36 nM) this activity did not correlate with the potency of inhibition of megakaryocyte development. Furthermore, other PDEIII inhibitors of widely differing potency were shown to have negligible effects on megakaryocytopoiesis.

6 Taken together our results demonstrate that anagrelide and BCH24426 target a cellular event involved specifically in the megakaryocyte differentiation programme, which is independent of PDEIII inhibition.
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Abbreviations: ANOVA, analysis of variance; BCH24426, 6,7-dichloro-3-hydroxy-1,5 dihydro-imidazo[2,1-b]quinazolin-2-one; CD, cluster differentiation antigen number; DMSO, dimethyl sulfoxide; EPO, erythropoietin; FITC, fluorescein isothiocyanate; GM-CSF, granulocyte macrophage colony-stimulating factor; GpA, glycophorin A; IBMX, isobutyl-1-methylxanthine; IMDM, Iscove's-modified Dulbecco's medium; MK, megakaryocyte; PBS, phosphate-buffered saline; PDEIII, phosphodiesterase type III; RL603, 2-amino-5,6-dichloro-3,4,-dihydroquinazoline; SDF-1α, stromal cell-derived factor-1α; TPO, thrombopoietin

Introduction

Anagrelide is an imidazoquinazoline derivative that was initially developed as an inhibitor of platelet aggregation (Fleming & Buyniski, 1979). However, when first tested in humans it showed profound thrombocytopenic effects (Abe *et al.*, 1984). This unexpected action prompted evaluation of the possible clinical utility of this property, which ultimately demonstrated the usefulness of the drug for the treatment of thrombocytosis in patients with chronic myeloproliferative disorders (reviewed in Pescatore & Lindley, 2000). Initial studies in humans indicated that anagrelide does not significantly affect platelet-survival time (Abe *et al.*, 1984) nor that it acts by inducing haematopoietic progenitor cell toxicity (Silverstein *et al.*, 1988). Later on, *in vitro* studies of human megakaryocytopoiesis suggested that, *in vivo*, the thrombocytopenic activity of this compound results primarily from an inhibitory effect on the postmitotic phase of megakaryocyte (MK) maturation (Mazur *et al.*, 1992). However, notwithstanding its clinical success and these pioneering cellular studies, unravelling the primary mechanism of action by which anagrelide reduces platelet count has remained elusive (Hong & Erusalimsky, 2002).

Anagrelide is extensively metabolized by the liver (Pescatore & Lindley, 2000). Early studies identified

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2-amino-5,6-dichloro-3,4,-dihydroquinazoline (RL603) as a major metabolite of the drug. Lane *et al.* (2001) investigated the activity of RL603 in MK cultures and also in the mouse. Their work suggested that this compound might be endowed with platelet-lowering activity, arising from inhibitory effects on MK maturation and migration. However, *in vitro* studies in our laboratories did not support this contention (Erusalimsky *et al.*, 2002). A second major metabolite of anagrelide, 6,7-dichloro-3-hydroxy-1,5 dihydro-imidazo[2,1-b]quinazolin-2-one, has recently been identified (Figure 1). Known as BCH24426, the activity of this metabolite has not been previously investigated.

To date the only known primary target of anagrelide is a type III phosphodiesterase (PDEIII) found in platelets and the myocardium (Gillespie, 1988; Beavo, 1995). While it would seem improbable that inhibition of this enzyme could account for the effects of anagrelide on MK development, this possibility has not been formally investigated.

To assess the potential role that the metabolites of anagrelide may play in the antithrombopoietic action of the parent compound in this study we have compared the activities of anagrelide, RL603 and BCH24426 for their effects on thrombopoietin (TPO)-induced haematopoietic progenitor $CD34^+$ cell expansion, as well as MK development and migration in culture. Furthermore, we have compared these compounds with other known PDEIII inhibitors in order to evaluate whether PDEIII inhibition could explain the anti-thrombopoietic activity of anagrelide.

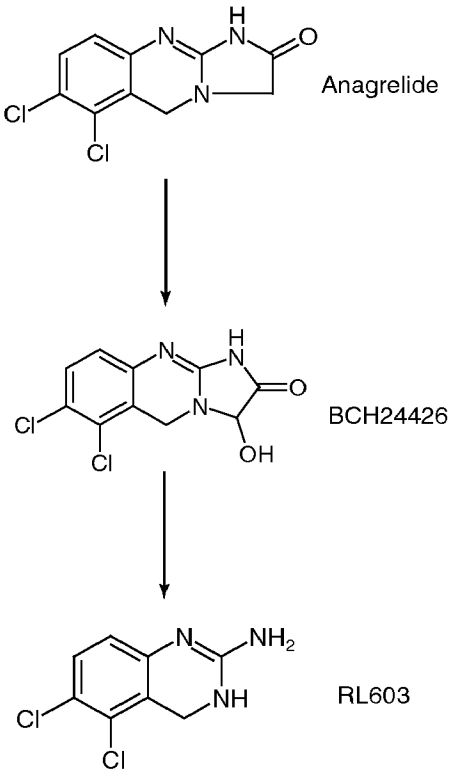

Figure 1 Metabolic pathway of anagrelide in man. Determined by mass spectrometric analysis of urine from volunteers given a single oral 1 mg dose of [^{14}C]-labelled anagrelide.

Methods

Chemicals

Anagrelide hydrochloride monohydrate (Batch No CML-227/01-RS6, >99% purity) was obtained from Cambridge Major Laboratories Inc., Germantown, WI, U.S.A. BCH24426 (Batch No MSC114/15, >94% purity) was synthesized by Tocris Cookson, Bristol, U.K. RL603 was obtained from Ultrafine, Manchester, U.K. (Batch No 116-44-1, 98% purity) or from York Bioanalytical Solutions, Upper Poppleton, U.K. (Batch No 130-24-1, purity ~100%). Cilostazol was obtained from Sigma-Aldrich, Poole, U.K. Trequinsin hydrochloride, milrinone and cilostamide were purchased from Calbiochem, Nottingham, U.K. Isobutyl-1-methylxanthine (IBMX) was from Sigma, Poole, U.K. Trequinsin hydrochloride was dissolved in water. Stocks solutions of RL603 were made in dimethyl sulfoxide (DMSO) or in acidic (pH 5.0) phosphate-buffered saline (PBS). All other test compounds were dissolved in DMSO. Stock solutions (10 mM) were stored at −20°C in small aliquots and when required, diluted in culture medium immediately before addition to cell suspensions.

Cells

Umbilical cord blood was obtained from normal donors undergoing scheduled caesarean births and who gave informed written consent in accordance to University College London Ethics Committee guidelines. $CD34^+$ haematopoietic progenitor cells were isolated by magnetic immunoselection using the anti-CD34 monoclonal antibody QBEND/10 (Miltenyi Biotec Ltd, Surrey, U.K.) as previously described (Mathur *et al.*, 2004). Cell purity was checked by flow cytometry and ranged between 92 and 98%.

Cell culture and treatments with test compounds

To examine the effect of test compounds on the expansion of $CD34^+$ cells resulting from TPO stimulation, these cells were grown under conditions which support the proliferation of MK progenitors in serum-free medium (Lam *et al.*, 2001; van den Oudenrijn *et al.*, 2001). Freshly isolated cells were cultured with 40 ng ml^{-1} recombinant human TPO (R&D Systems, Abingdon, U.K.) in Stemspan™ medium (Stem Cell Technologies, London, U.K.) which consisted of Iscove's-modified Dulbecco's medium (IMDM) supplemented with 1% bovine serum albumin, 10 μg ml^{-1} recombinant human insulin, 200 μg ml^{-1} iron-saturated transferrin, 0.1 mM 2-mercaptoethanol and 2 mM glutamine. Cells were seeded onto flat bottom 96-well microtitre plates (Falcon) at an initial density of 1.5×10^5 cells ml^{-1} in a volume of 100 μl and incubated at 37°C in a humidified incubator under 5% CO_2/95% air. At 1 day after the initial seeding, test compounds or vehicle were added at the indicated doses in a volume of 10 μl and the cells were cultured for a further 5 days. At the end of the culture period the relative number of viable cells was determined using a tetrazolium salt-based colourimetric assay (XTT, Cell proliferation kit II, Roche Diagnostics GmbH, Mannheim, Germany) as described by the manufacturers.

To promote terminal differentiation, unless otherwise indicated, freshly isolated $CD34^+$ cells were cultured in

standard differentiation medium consisting of IMDM (Sigma) supplemented with 10% human umbilical cord blood plasma, 0.2% bovine serum albumin, 2 mM glutamine, 1 mM sodium pyruvate, 0.1 mM nonessential amino acids (Gibco BRL), minimal essential medium vitamins (Gibco BRL), 0.1 mM 2-mercaptoethanol, 100 U ml^{-1} penicillin, 0.1 mg ml^{-1} streptomycin and specified haematopoietic growth factors. In some experiments cells were cultured in the above-described Stemspan™ serum-free medium. Cells were grown for 12 days at 37°C in a humidified incubator under 5% CO_2/95% air. The final cell density of the differentiated cultures was determined using a Sysmex CDA-500 Particle Analyser fitted with a 5–20 μm probe. Treatments with test compounds under differentiation conditions were carried out according to two alternative schedules. Schedule A was used to evaluate the activity of test compounds on the growth and differentiation of developing MKs. In this case cells were plated at a density of 2.0×10^5 cells ml^{-1} in medium containing 40 ng ml^{-1} TPO. After 4 days of culture, cells were counted and then diluted ($\sim$3-fold) to 1.5×10^5 cells ml^{-1} by addition of fresh medium supplemented with 10 ng ml^{-1} TPO. Following dilution, cell aliquots of 1 ml were replated into 24-well plates and test compounds or vehicle were added at the appropriate doses in a volume of 20 μl. Cells were then left to grow undisturbed until the end of the 12-day culture period. Schedule B was used to compare the activity of test compounds against the differentiation of $CD34^+$ cells into different lineages as follows: Cell aliquots of 1 ml were seeded onto 24-well plates at a density of 1.5×10^5 cells ml^{-1} with medium containing either 40 ng ml^{-1} TPO, 8 U ml^{-1} recombinant human erythropoietin (EPO, Roche Diagnostics GmbH) or 20 ng ml^{-1} recombinant human granulocyte–macrophage colony-stimulating factor (GM-CSF, R&D Systems). After an overnight incubation, drugs or vehicle were added as described above. Subsequently, the cells were left to grow undisturbed until the end of the 12-day culture period.

Flow cytometric analysis of cell differentiation

Phenotypic differentiation was monitored by flow cytometry using the following fluorescein isothiocyanate (FITC)-conjugated monoclonal antibodies: Y2/51 (DAKO, U.K.), which detects the megakaryocytic lineage-specific marker CD61; CLB-409 (Cymbus Biotechnology, Hants, U.K.), which recognizes the erythroid differentiation marker glycophorin A (GpA) and MΦP9 (Becton Dickinson, U.K.), which detect the antigen CD14 on myelomonocytic cells. Cells were stained and analysed as previously described (Bobik *et al.*, 1998; Mathur *et al.*, 2004). The boundary between antigen-positive and -negative cells was established according to the fluorescence distribution of cells stained with an isotype-matched control antibody. The number of differentiated cells was estimated by multiplying the total number of cells in the culture by the percentage of antigen-positive cells. Relative cell size was determined by flow cytometry from the mean forward scatter signal of the cell population. Determination of DNA ploidy distributions in megakaryocytic cultures was carried out by double labelling with FITC-conjugated Y2/51 and propidium iodide (Sigma) as previously described (Mathur *et al.*, 2004).

Transmigration assay

Transmigration experiments were carried out using a bicompartmental system (Costar, U.K.) consisting of a Transwell® chamber (5 μm membrane pore size) inserted onto the well of a 24-well tissue culture plate essentially as we have previously described (Mathur *et al.*, 2001). Experiments were performed using unfractionated terminally differentiated MK cultures (1.0 1.5×10^6 cells ml^{-1}) that had been previously grown in standard differentiation medium containing 40 ng ml^{-1} TPO as described above. The proportion of $CD61^+$ cells in these cultures ranged between 65 and 80%. Cell suspensions were used in their own conditioned medium without further processing. Aliquots of the cell suspension (0.1 ml) were placed onto the Transwell and test compounds, or an equivalent amount of vehicle, were added to give a final concentration of 1 μM. The lower compartment was filled with fresh 0.6 ml standard differentiation medium with or without 150 ng ml^{-1} stromal cell-derived factor 1α (SDF-1α, R&D Systems). Plates were placed in an incubator at 37°C under 5% CO_2 for 4 h. At the end of this period the Transwell insert was removed and migrated cells were collected from the bottom well. Both the bottom well and the external surface of the insert were rinsed with 1 ml PBS containing 2 mM EDTA to ensure that all cells were collected. SDF-1α-induced MK migration was quantified by flow cytometry following staining of the collected cells with FITC-conjugated Y2/51 monoclonal antibody as described above. Each sample was resuspendend in a final volume of 0.4 ml and cells were counted twice using the flow cytometer time parameter, which was set to acquire events for 30 s. Analysis gates were established to assess the migration of CD61-positive and -negative cells. Regular checks were made to ensure that the volume of cells processed by the instrument during the 30-s acquisition period remained constant.

Data processing and statistical analysis

Each experiment was carried out in 2–3 replicates with cells derived from the same donor and the results were averaged. Experiments were repeated at least three times using each time $CD34^+$ cells from a different donor. To account for variations in the differentiation responses between different batches of $CD34^+$ cells, results were normalized to percentages of control (no drug added) and then averaged. Statistical analysis was carried out using SPSS software (release 12, SPSS Inc., Chicago, IL, U.S.A.). A two-tailed Student's *t*-test was used to establish levels of significance for differences between two values. For comparing dose–responses levels of significance were determined by two-way analysis of variance (ANOVA). Differences resulting in $P<0.05$ were considered significant. IC_{50} values were calculated using Prism software (release 3.0, GraphPad Software Inc., San Diego, CA, U.S.A.).

Results

Activity of anagrelide and its metabolites against the early expansion of $CD34^+$ cell cultures stimulated by TPO

Figure 2 shows the dose–responses for the effects of anagrelide, RL603 and BCH24426 on the early expansion of

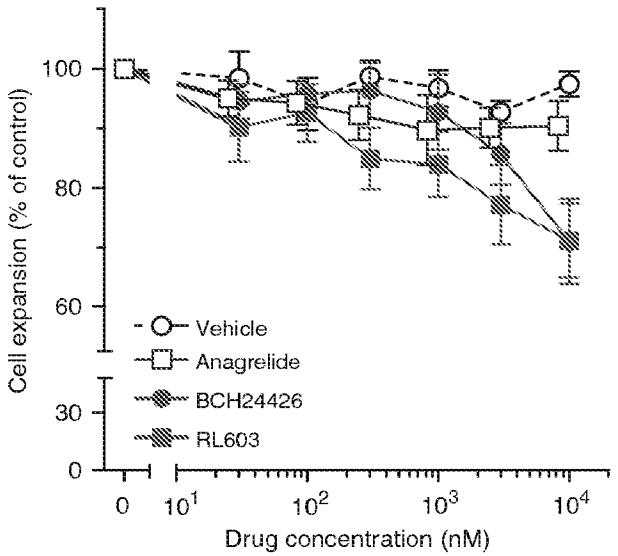

Figure 2 Dose–responses for the effects of anagrelide and its metabolites on TPO-induced CD34$^+$ cell expansion. CD34$^+$ cells were cultured for 6 days in Stemspan™ serum-free expansion medium supplemented with TPO. The indicated compounds or an equivalent amount of vehicle (DMSO) were added after 24 h. Cell expansion was determined by the XTT assay and is expressed relative to untreated samples grown in parallel. Results represent the mean ± s.e.m. of three to five experiments.

CD34$^+$ cell cultures grown with TPO in serum-free medium. Under these experimental conditions (6 days of culture), control cells underwent 6.9 ± 1.9-fold expansion ($n = 5$). Each of the test compounds had weak inhibitory activity against this process (5–15% inhibition at 1 μM, reaching 10–30% at 10 μM). Of the three compounds anagrelide appeared to be the weakest (ANOVA, $P = 0.06$ vs vehicle), whereas RL603 and BCH24426 showed stronger comparable activity (ANOVA, $P < 0.01$ for each treatment vs vehicle). However, due to the variability in the responses only the latter demonstrated a significant effect of dose (ANOVA, $P = 0.02$ vs vehicle) and this was accounted exclusively by a reduction in the number of viable cells at the highest concentration tested (~30% inhibition at 10 μM BCH24426, $P = 0.004$ vs vehicle by t-test). Furthermore, comparison between the dose–responses of anagrelide and BCH24426 showed the two compounds to be indistinguishable (ANOVA, $P = 0.12$).

Differential effects of anagrelide metabolites against megakaryocytopoiesis

To compare the effects of anagrelide and its metabolites on the *ex vivo* production of MKs, CD34$^+$ cells were grown in differentiation medium supplemented with TPO and test compounds were added after 1 or 4 days of culture (schedules B and A, respectively). Preliminary experiments demonstrated that there was no substantial difference in the final effect of the test compounds using these alternative schedules (data not shown). However, because schedule A allows for the expansion of the MK progenitors before they are exposed to the test compounds, a much large number of samples can be tested in parallel under these conditions. Hence schedule A was used in most of the subsequent experiments in which MK development was examined. Figure 3 shows representative flow cytometric profiles of CD61 expression (a measure of differentiation) after 12 days of culture. In control cultures, the majority of cells expressed very high relative levels of this megakaryocytic differentiation marker (~100-fold above those of the negative fraction). Addition of anagrelide or BCH24426 reduced the fraction of CD61^{bright} cells in a dose-dependent manner, with a maximal effect observed at ~1 μM (Figure 3 and data not shown). In contrast, flow cytometric profiles of RL603-treated cultures were virtually indistinguishable from those of control cultures (Figure 3a and data not shown). As shown on Table 1 at a concentration of 1 μM

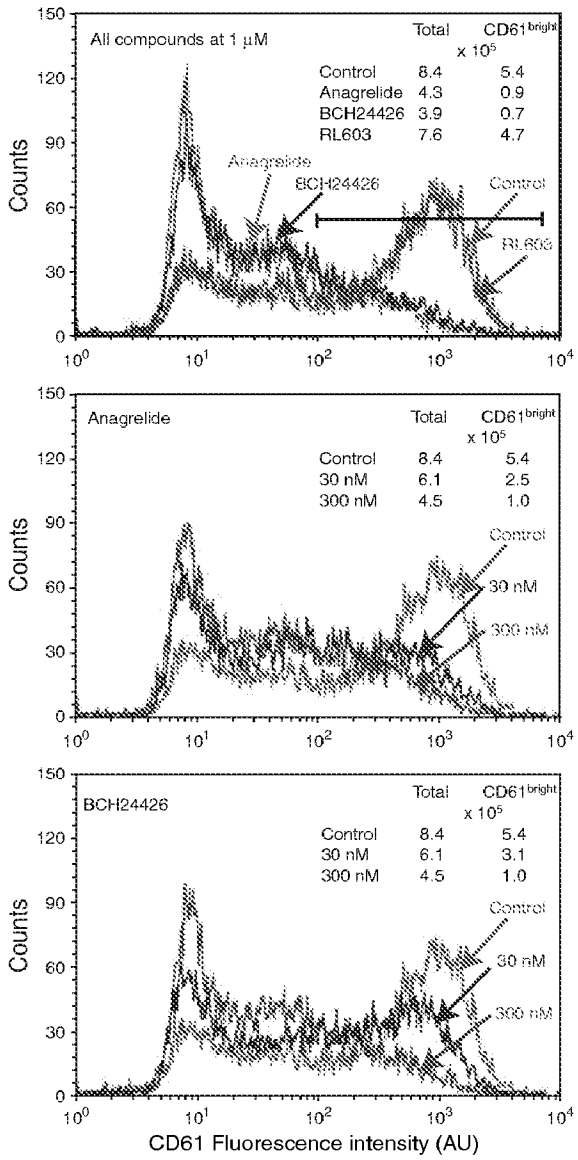

Figure 3 Flow cytometric histograms of CD61 expression in cultured MKs treated with anagrelide or its metabolites. CD34$^+$ cells were cultured for 12 days in differentiation medium supplemented with TPO in the presence or absence of the indicated compounds as described in schedule A under Methods and then analysed by flow cytometry. The marker encompasses the CD61^{bright}-positive cells. The inset tables show both the total number of cells and the number of CD61^{bright}-positive cells in the respective cultures.

Table 1 Effects of anagrelide and its metabolites on the growth characteristics of MK cultures

Treatment	*Cell density*[a] *(%)*	*Relative cell size*[a] *(%)*	*CD61 expression* *CD61*^{bright} *cell fraction*[a] *(%)*	*Median CD61 fluorescence*[b] *(%)*
Vehicle	95.8±2.2	105.5±3.0	99.1±1.6	98.5±3.4
Anagrelide	48.0±4.0**	80.4±4.5**	41.9±6.6**	51.3±11.9**
BCH24426	45.8±3.9**	78.7±5.3**	34.5±4.4**	54.4±14.7*
RL603	93.9±7.2	103.1±4.1	94.2±2.2	90.2±13.3

$CD34^+$ cells were cultured in differentiation medium supplemented with TPO in the presence or absence of the indicated compounds (1.0 μM) or an equivalent amount of vehicle (0.01% DMSO) as described in Figure 3 and then analysed by flow cytometry. Results are expressed in percentages relative to untreated cells grown in parallel. Values represent the mean±s.e.m. of four experiments. *$P<0.05$; **$P<0.01$ vs vehicle.
[a]Values correspond to the total cell population.
[b]Values correspond to the $CD61^{bright}$ cell fraction.

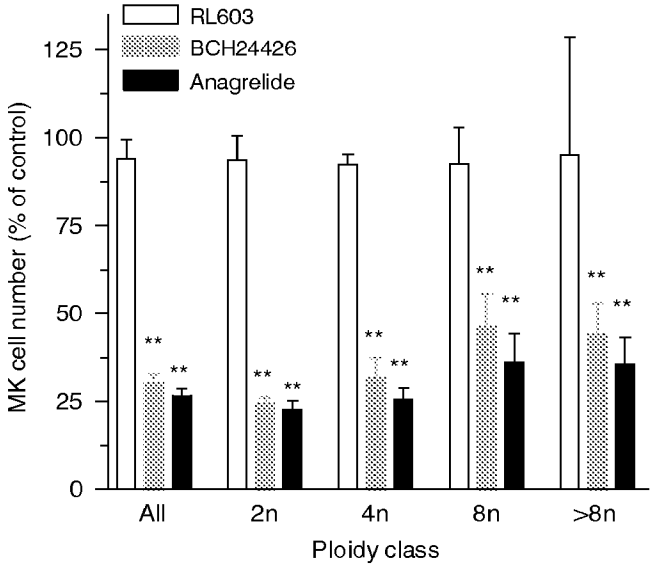

Figure 4 Effects of anagrelide and its metabolites on the ploidy distribution of cultured MKs. $CD34^+$ cells were cultured with the indicated compounds (1.0 μM) as described in Figure 3. Results show the total number of $CD61^{bright}$ cells and the number in each ploidy class relative to untreated samples grown in parallel. Values represent the mean±s.e.m. of three or four experiments. **$P<0.01$ vs vehicle.

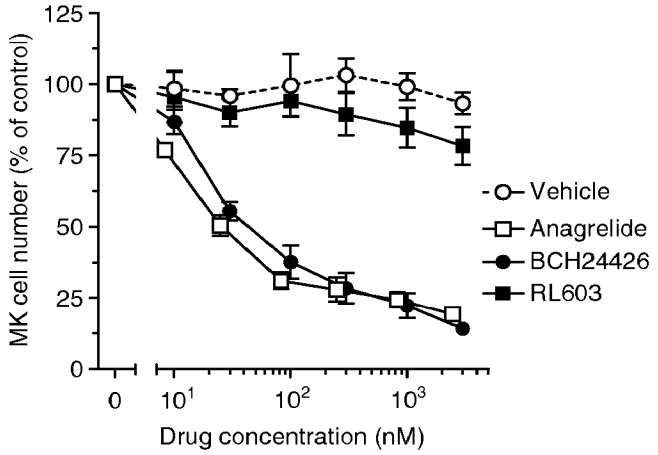

Figure 5 Dose–responses for the effects of anagrelide and its metabolites on TPO-induced MK development. $CD34^+$ cells were cultured with the indicated compounds as described in Figure 3. Results show the total number of $CD61^{bright}$ cells relative to untreated samples grown in parallel. Values represent the mean± s.e.m. of eight experiments.

anagrelide and BCH24426 reduced the fraction of positive cells by an average of 60–65%. In addition, these compounds reduced significantly the median fluorescence intensity of the $CD61^{bright}$ subpopulation (a measure of the relative level of antigen expression per cell), the average cell size of the whole culture (the latter is a function of both cytoplasmic maturation and DNA content) and its final cell density. In contrast, RL603 had no significant effect on any of these variables (Table 1). Furthermore, flow cytometric analysis of ploidy distributions showed that anagrelide and BCH24426 reduced the proportion of MKs in each ploidy class to similar extents whereas RL603 again showed no significant effect (Figure 4).

Figure 5 shows the dose–responses for the overall activity of anagrelide and its metabolites against *in vitro* megakaryocytopoiesis as assessed by evaluating their effects on the total number of MKs ($CD61^{bright}$ cells) produced in the culture. These results show that anagrelide and BCH24426 inhibit MK development with high efficacy and very similar potencies (mean IC_{50}±s.e.m. = 26±4 and 44±6 nM, respectively; ANOVA, $P=0.8$ between compounds). In contrast, in the same concentration range RL603 showed only weak activity and no significant effect of dose (ANOVA, $P=0.5$ compared to vehicle); hence in this case an IC_{50} value could not be established.

The poor effect of RL603 on MK development compared to the strong inhibition of this process by anagrelide or BCH24426 seen in the above described experiments was reproduced when cultures were grown for 12 days in serum-free medium, when RL603 was prepared in acidic PBS or when material from an alternative supplier was tested (data not shown).

Selective inhibition of megakaryocytopoiesis by anagrelide and BCH24426

To assess whether the inhibitory activities of anagrelide and BCH24426 in these cultures were selective for the megakaryocytic lineage, we examined the effects of these compounds on the growth of the nonmegakaryocytic cells ($CD61^-$). In 12-day control cultures, these cells represent 20–35% of the total

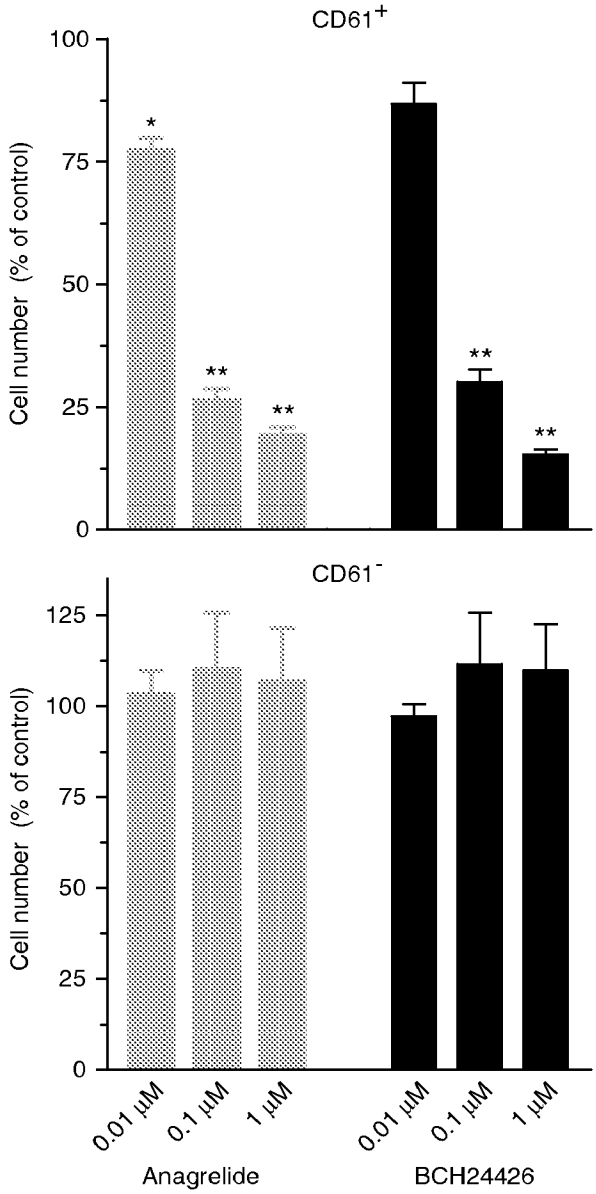

Figure 6 Effect of anagrelide and BCH24426 on the growth on nonmegakaryocytic cells. $CD34^+$ cells were cultured with the indicated compounds as described in Figure 3. Results show the total number of $CD61^-$ and $CD61^+$ cells relative to untreated samples grown in parallel. Values represent the mean±s.e.m. of four experiments. $^*P<0.05$; $^{**}P<0.01$ vs vehicle.

population (Figure 3). As depicted in Figure 6, in sharp contrast to the reduction in the final number of cells expressing CD61, neither anagrelide nor BCH24426 inhibited the growth of nonmegakaryocytic cells.

To obtain further evidence for the selectivity of these compounds their effects on the differentiation of CD34 cells into other haematopoietic lineages was also tested. As shown in Figure 7, neither anagrelide nor its metabolites had a significant effect on erythroid or myelomonocytic differentiation induced by EPO or GM-CSF. In contrast, in the same experiments, both anagrelide and BCH24426 had a strong inhibitory effect on megakaryocytic differentiation induced by TPO.

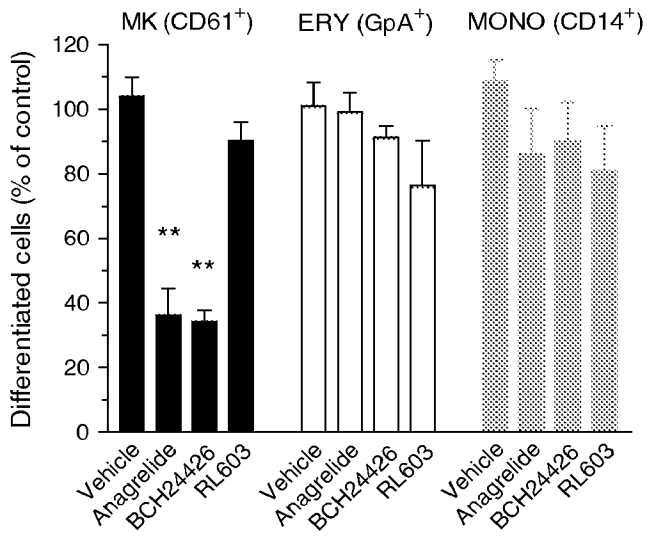

Figure 7 Comparison between anagrelide and its metabolites for their effects on haematopoietic lineage differentiation. $CD34^+$ cells were cultured for 12 days in differentiation medium supplemented with TPO, EPO or GM-CSF in the presence or absence of the indicated compounds (1.0 µM) or an equivalent amount of vehicle (0.01% DMSO) as described in schedule B under Methods. Results show the total number of megakaryocytic (MK, $CD61^+$), erythroid (ERY, GpA^+) and myelomonocytic (MONO, $CD14^+$) cells relative to untreated samples grown in parallel. Values represent the mean±s.e.m. of four experiments. $^{**}P<0.01$ vs vehicle.

Lack of activity of anagrelide and its metabolites against SDF-1α-stimulated MK migration

Figure 8 shows the results of a representative experiment in which anagrelide and its metabolites were tested for their effects on the migration of cells derived from $CD34^+$ cell cultures. Both $CD61^+$ megakaryocytic cells and $CD61^-$ nonmegakaryocytic cells exhibited a strong migratory response to the chemotactic factor SDF-1α. However, neither anagrelide nor its metabolites showed a significant effect on the migratory response. It has been suggested that preparation of RL603 in acidic aqueous buffers increases its solubility and hence its activity (Rafii & Lane, 2002). However, as shown in Figure 8, when RL603 was prepared in this manner it still failed to inhibit SDF-1α-induced migration.

In order to confirm these findings experiments were repeated several times using cells from different donors. As shown in Table 2, on average all the compounds caused a small inhibition of SDF-1α-dependent MK migration. However, none of the effects reached statistical significance.

Evidence that phosphodiesterase inhibition is not related to the antimegakaryocytopoietic activity of anagrelide and BCH24426

As shown in Table 3, consistent with previous reports (Gillespie, 1988) anagrelide was found to inhibit PDEIII *in vitro*. Furthermore, BCH24426 was found to be 40-fold more potent than anagrelide against this activity. These findings contrasted with the fact that both compounds had similar potencies when tested for their effects on megakaryocytopoiesis (Figure 5) and thus suggest that the mechanism by which they abrogate this process is unrelated to PDEIII inhibition.

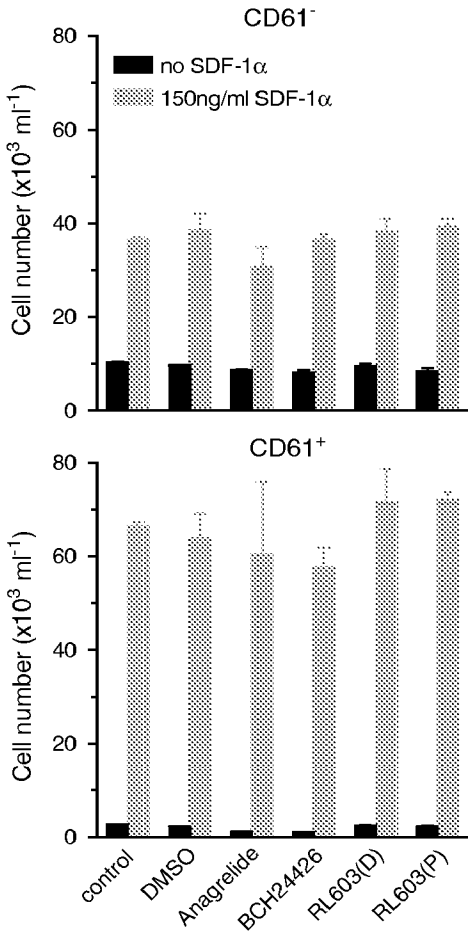

Figure 8 Effects of anagrelide and its metabolites on the transmigration of $CD61^+$ and $CD61^-$ cells. Transmigration experiments were carried out in the presence or absence of the indicated compounds (1.0 μM) or 0.01% DMSO as described under Methods. Results represent the mean±s.e.m. number of migrated cells for three determinations in one representative experiment. RL603 was dissolved in DMSO (D) or in acidic PBS (P).

In order to substantiate this contention, further experiments were carried out with a number of commercially available PDEIII inhibitors. As summarized in Table 3 while both anagrelide and BCH24426 showed strong and significant inhibitory effects against MK development, apart from trequinsin (*t*-test, $P=0.04$ vs untreated cells), none of the other PDEIII inhibitors showed a statistically significant effect against this process. In the case of trequinsin, however, the overall effect was small, even though the compound was used at a concentration equivalent to 3000-fold its IC_{50} for PDEIII.

Discussion

In these investigations we have examined the activity of two major metabolites of anagrelide, RL603 and BCH24426, against several *in vitro* parameters related to the process of megakaryocytopoiesis and platelet production *in vivo*. These parameters included the early expansion of undifferentiated MK progenitors, the degree of MK maturation (relative cell size, ploidy and level of CD61 expression), the overall number

Table 2 Effects of anagrelide and its metabolites on MK migration

Treatment	*SDF-1α-dependent MK migration (%)*	P*
DMSO, 0.01%, $n=5$	98.0±7.9	
Anagrelide, $n=5$	83.3±10.3	0.29
BCH24426, $n=5$	74.2±8.9	0.08
RL603 (D)[a], $n=5$	83.6±8.3	0.25
RL603 (P)[a], $n-3$	80.4±18.2	0.29

Transmigration experiments were carried out as described in Figure 8. Results show the total number of $CD61^+$ cells that migrated towards the SDF-1α gradient, expressed as a percentage of cells migrating towards the gradient in the absence of test compound. Values represent the mean±s.e.m. of the indicated number experiments.
*P values were calculated by a *t*-test; RL603 (P) was compared to untreated cells and the remaining compound treatments were compared to DMSO.
[a]The letters in brackets denote whether RL603 was dissolved in DMSO (D) or acidic PBS (P).

Table 3 Comparison between PDEIII inhibitors for their activity against megakaryocytopoiesis

Treatment	*PDEIII activity IC_{50} (nM)*	*MK cell number[a] (%)*
DMSO, 0.01%	—	91.8±7.3
Trequinsin	0.3[b]	83.5±5.3[#]
BCH24426	0.9[c]	27.5±3.9**
Cilostamide	27[b]	86.3±7.1
Anagrelide	36[c]	26.1±2.7**
Cilostazol	200[b]	79.1±8.0
Milrinone	450[b]	94.6±7.1
IBMX	3950[b]	85.8±8.6

$CD34^+$ cells were cultured in differentiation medium supplemented with TPO in the presence or absence of the indicated compounds (1.0 μM) or an equivalent amount of DMSO as described in schedule A under Methods.
[a]The number of MKs in the cultures is shown in percentages relative to untreated cells; values represent the mean±s.e.m. of three experiments.
[b]IC_{50} values for trequinsin (Ruppert & Weithmann, 1982), Cilostamide (Sudo *et al.*, 2000), cilostazol (Sudo *et al.*, 2000), milrinone (Sudo *et al.*, 2000) and IBMX (Tang *et al.*, 1994) were obtained from the literature.
[c]PDEIII activity was measured by assessing the production of 5′-AMP from cAMP using enzyme isolated from human platelets.
[#]$P<0.05$ vs untreated cells; **$P<0.01$ vs DMSO.

of CD61-expressing cells and the migration of MKs stimulated by SDF-1α. Our results demonstrate that BCH24426 is a potent inhibitor of megakaryocytopoiesis, with virtually identical effects to those observed with anagrelide, in terms of potency, efficacy, mode of action and specificity. These conclusions are based on the following findings: Firstly, anagrelide and BCH24426 affected the same MK differentiation parameters and to similar extents (Table 1, Figures 3 and 4). Secondly, both drugs showed similar IC_{50} values for their overall effect on the generation of differentiated MKs (Figure 5). Thirdly, in the 0.1–1 μM concentration range, that is, at concentrations that effectively inhibited MK development, neither compound showed a substantial effect against the TPO-induced early expansion of progenitor cells (Figure 2), against the growth of nonmegakaryocytic cells (Figure 6), against the differentiation of haematopoietic progenitor cells

into other lineages (Figure 7) or against MK migration (Figure 8). Thus, taken together these findings suggest that both anagrelide and BCH24426 act selectively on the growth and maturation of committed MK precursors.

Cell culture studies carried out by another laboratory had previously suggested that RL603 is 50 times more potent than anagrelide as an inhibitor of MK maturation and that this compound also inhibits MK migration (Lane *et al.*, 2001). In preliminary studies (Erusalimsky *et al.*, 2002), which we have now extended by the present work, we could not confirm those claims. Indeed, while we found some evidence that RL603 has antimegakaryocytopoietic activity, the overall inhibition was small, even at relatively high doses (Figure 5). Furthermore, we have not found any evidence that RL603 inhibits MK migration to a significant extent (Figure 8 and Table 2). Rafii & Lane (2002) have previously indicated that in order to increase the solubility of RL603, the compound ought to be prepared in acidic aqueous media and/or that the presence of DMSO may interfere with the sensitivity of MK development bioassays. However, in the present work we could not corroborate those claims. Indeed, in our hands the activity of RL603 did not improve when the drug was prepared in acidic aqueous media and when there were no traces of DMSO in the assay. Another possibility is that differences in experimental culture conditions accounted for these contradictory findings. Rafii & Lane (2002) carried out their *in vitro* studies in plasma-free medium. However, in our studies the use of plasma-free or plasma-containing media did not affect the activity of RL603. Consistent with these *in vitro* findings, *in vivo* studies conducted by one of the authors failed to show any effect of RL603 on platelet counts when administered to rats or mice (Franklin, unpublished data). Thus, an explanation for these apparently conflicting data on the activity of RL603 would require further experimental work.

A salient finding of the present study is that anagrelide and BCH24426 inhibited MK development at low nanomolar concentrations (Figure 5). In the case of anagrelide these doses are well within the range of therapeutic concentrations reached in the human circulation. For instance, after administration of a typical 1 mg oral dose to patients with essential thrombocythaemia the mean maximum plasma concentration was $6.2\,\mathrm{ng\,ml^{-1}} = 24\,\mathrm{nM}$ (data on file at Shire Pharmaceuticals). In the case of BCH24426 the corresponding mean maximum plasma concentration was $8.7\,\mathrm{ng\,ml^{-1}} = 32\,\mathrm{nM}$, which again lies within the range observed to be effective in the current *in vitro* studies.

Previous findings have suggested that anagrelide acts primarily on the postmitotic phase of MK maturation (Mazur *et al.*, 1992; Solberg *et al.*, 1997). More recently work by another laboratory has shown that *in vivo* anagrelide also reduces MK cell numbers (Tomer, 2002). Consistent with this finding our results in liquid cultures show that nanomolar concentrations of anagrelide and BCH24426 not only affect the degree of maturation (Figure 3 and Table 1) but also caused a dose-dependent decline in the overall number of MKs (Figure 5). This reduction in MK cell number could not be attributed to indiscriminate cytotoxic effects, as the compounds did not reduce the number of nonmegakaryocytic cells in cultures grown with TPO (Figure 6), nor did they significantly affect the number of erythroid or myelomonocytic cells in cultures grown with EPO and GM-CSF, respectively (Figure 7). Furthermore, the lack of significant interference with the expansion of early MK progenitors at concentrations, which effectively inhibited overall MK development, that is, in the 0.1–1.0 μM range (compare Figure 2 with 5), suggest that anagrelide and BCH24426 inhibit proliferation at a late stage in the mitotic phase of megakaryocytopoiesis, that is, once a substantial degree of differentiation has taken place. This possibility is entirely consistent with the notion that there is a considerable degree of overlap between the proliferative and maturation phases of megakaryocytopoiesis (Long, 1993; Hong & Erusalimsky, 2002).

It could be argued that the reduction in MK numbers is due to the active compounds having a selective toxic effect on the developing MK subpopulation. However, this possibility is unlikely because effective concentrations of anagrelide did not increase the basal level of cell death in these cultures (Hong & Erusalimsky, manuscript in preparation).

Anagrelide inhibits PDEIII found in platelets and as a result raises cAMP levels in these cells (Gillespie, 1988; Seiler *et al.*, 1987). This action may explain its inhibitory effect on platelet aggregation. Although after *in vivo* administration of doses which are effective in reducing blood platelets, this activity is very short lived, the role of PDEIII inhibition in the thrombocytopenic action of the drug has never been entirely discounted. BCH24426 like anagrelide inhibits PDEIII *in vitro*, the former being 40 times more potent. These wide differences in potency with regards to PDEIII inhibition but not with regards to megakaryocytopoiesis, and the fact that a number of other known phosphodiesterase inhibitors did not have any effect on the latter (Table 3) strongly suggests that PDEIII inhibition is not the mechanism by which anagrelide inhibits platelet production. In conclusion, our data indicate that anagrelide and BCH24426 target a cellular event involved specifically in the megakaryocytic differentiation programme. These findings suggest that *in vivo* BCH24426 could also play a role in the platelet-lowering action of the parental compound.

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